US007989429B2

(12) **United States Patent**
Worm

(10) **Patent No.: US 7,989,429 B2**
(45) **Date of Patent: Aug. 2, 2011**

(54) **LNA ANTAGONISTS TARGETING THE ANDROGEN RECEPTOR**

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(73) Assignees: **Enzon Pharmaceuticals, Inc.**, Bridgewater, NJ (US); **Santaris Pharma A/S**, Hoersholm (DK)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

(21) Appl. No.: **12/726,554**

(22) Filed: **Mar. 18, 2010**

(65) **Prior Publication Data**

US 2010/0234451 A1 Sep. 16, 2010

Related U.S. Application Data

(63) Continuation of application No. 12/324,033, filed on Nov. 26, 2008, now Pat. No. 7,737,125.

(60) Provisional application No. 60/990,125, filed on Nov. 26, 2007.

(51) **Int. Cl.**
A61K 48/00 (2006.01)
C07H 21/04 (2006.01)

(52) **U.S. Cl.** ... **514/44**; 536/23.1; 536/24.31; 536/24.33; 536/24.5

(58) **Field of Classification Search** ..... None
See application file for complete search history.

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Beane et al., Inhibiting gene expression with locked nucleic acids (LNAs) that target chromosomal DNA. Biochemistry vol. 46, pp. 7572-7580, 2007.

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Primary Examiner — Amy Bowman
(74) *Attorney, Agent, or Firm* — Lucas & Mercanti, LLP

(57) **ABSTRACT**

The invention relates to oligonucleotide compounds (oligomers), which target androgen receptor mRNA in a cell, leading to reduced expression of the androgen receptor. Reduction of the androgen receptor expression is beneficial for the treatment of certain disorders, such as a hyperproliferative disorders (e.g., cancer). The invention provides therapeutic compositions comprising oligomers and methods for modulating the expression of androgen receptor using said oligomers, including methods of treatment.

10 Claims, 22 Drawing Sheets

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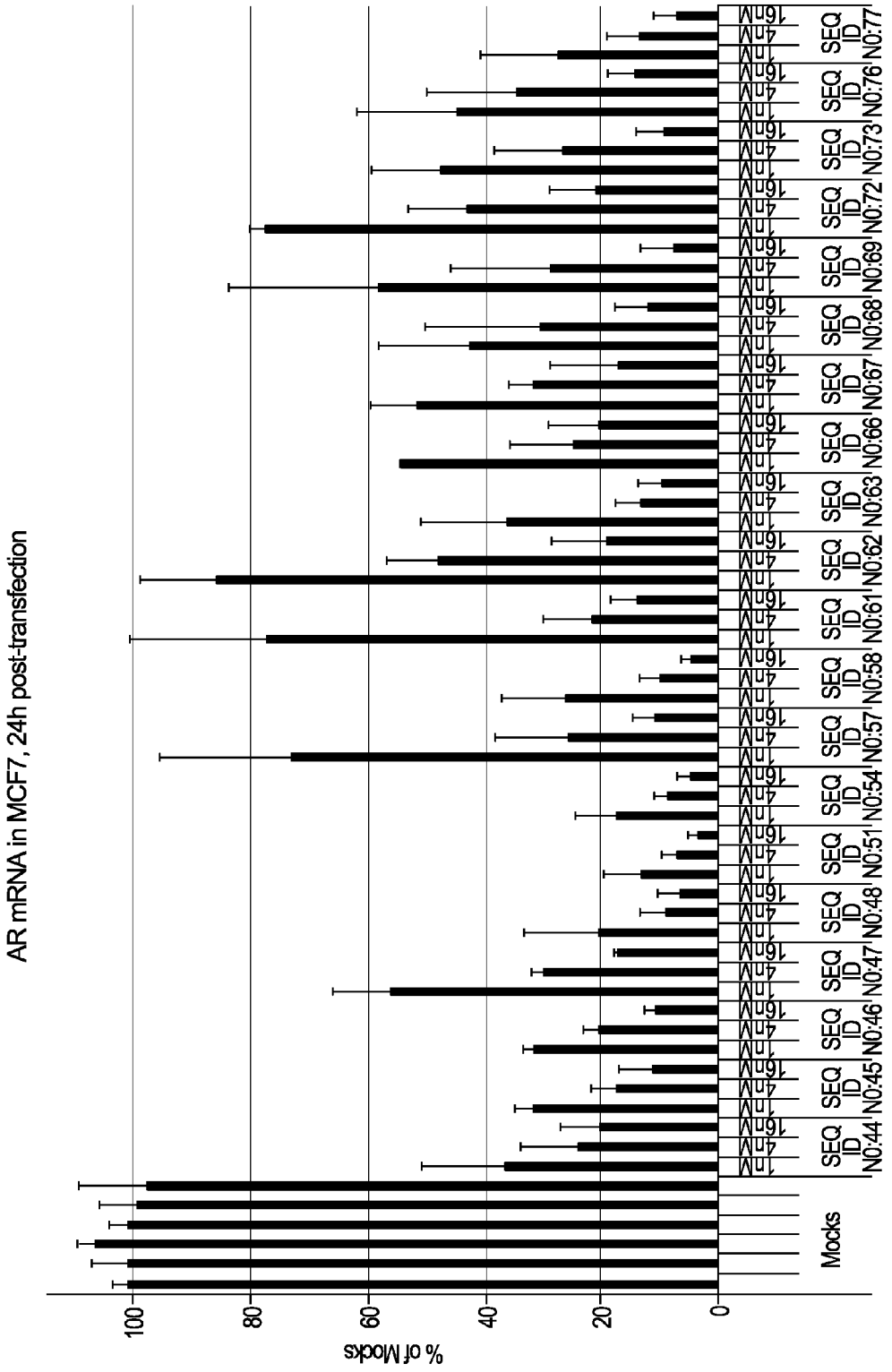
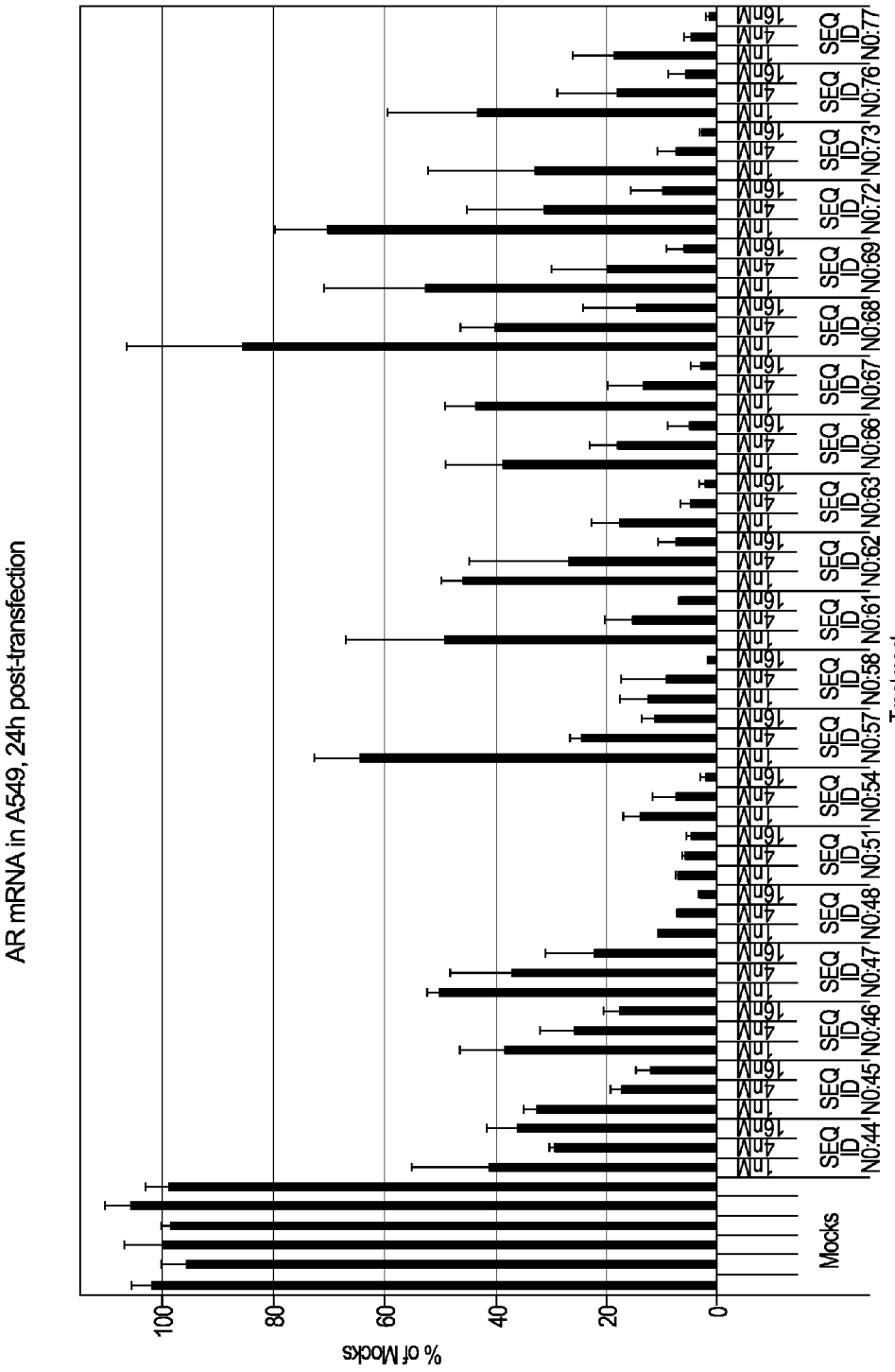

AR mRNA in MCF7, 24h post-transfection
% of Mocks
0
20
40
60
80
100
Mocks
SEQ ID NO:44
SEQ ID NO:45
SEQ ID NO:46
SEQ ID NO:47
SEQ ID NO:48
SEQ ID NO:51
SEQ ID NO:54
SEQ ID NO:57
SEQ ID NO:58
SEQ ID NO:61
SEQ ID NO:62
SEQ ID NO:63
SEQ ID NO:66
SEQ ID NO:67
SEQ ID NO:68
SEQ ID NO:69
SEQ ID NO:72
SEQ ID NO:73
SEQ ID NO:76
SEQ ID NO:77
4nM
16nM
Treatment

Figure 1


AR mRNA in A549, 24h post-transfection
% of Mocks
0
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40
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Mocks
SEQ ID NO:44
SEQ ID NO:45
SEQ ID NO:46
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SEQ ID NO:58
SEQ ID NO:61
SEQ ID NO:62
SEQ ID NO:63
SEQ ID NO:66
SEQ ID NO:67
SEQ ID NO:68
SEQ ID NO:69
SEQ ID NO:72
SEQ ID NO:73
SEQ ID NO:76
SEQ ID NO:77
4nM
16nM
Treatment

Figure 2

Figure 3

Alignment human and mouse AR mRNA

```
                    1                                                  50
NM_013476     (1)   ---------------------------------------------------
NM_000044     (1)   CGAGATCCCGGGGAGCCAGCTTGCTGGGAGAGCGGGACGGTCCGGAGCAA
Consensus     (1)
                    51                                                 100
NM_013476     (1)   ---------------------------------------------------
NM_000044     (51)  GCCCAGAGGCAGAGGAGGCGACAGAGGGAAAAGGGGCCGAGCTAGCCGCT
Consensus     (51)
                    101                                                150
NM_013476     (1)   ---------------------------------------------------
NM_000044     (101) CCAGTGCTGTACAGGAGCCGAAGGGACGCACCACGCCAGCCCCAGCCCGG
Consensus     (101)
                    151                                                200
NM_013476     (1)   ---------------------------------------------------
NM_000044     (151) CTCCAGCGACAGCCAACGCCTCTTGCAGCGCGGCGGCTTCGAAGCCGCCG
Consensus     (151)
                    201                                                250
NM_013476     (1)   ---------------------------------------------------
NM_000044     (201) CCCGGAGCTGCCCTTTCCTCTTCGGTGAAGTTTTTAAAAGCTGCTAAAGA
Consensus     (201)
                    251                                                300
NM_013476     (1)   ---------------------------------------------------
NM_000044     (251) CTCGGAGGAAGCAAGGAAAGTGCCTGGTAGGACTGACGGCTGCCTTTGTC
Consensus     (251)
                    301                                                350
NM_013476     (1)   ---------------------------------------------------
NM_000044     (301) CTCCTCCTCTCCACCCCGCCTCCCCCCACCCTGCCTTCCCCCCCTCCCCC
Consensus     (301)
                    351                                                400
NM_013476     (1)   ---------------------------------------------------
NM_000044     (351) GTCTTCTCTCCCGCAGCTGCCTCAGTCGGCTACTCTCAGCCAACCCCCCT
Consensus     (351)
                    401                                                450
NM_013476     (1)   ---------------------------------------------------
NM_000044     (401) CACCACCCTTCTCCCCACCCGCCCCCCCGCCCCCGTCGGCCCAGCGCTGC
Consensus     (401)
                    451                                                500
NM_013476     (1)   ---------------------------------------------------
NM_000044     (451) CAGCCCGAGTTTGCAGAGAGGTAACTCCCTTTGGCTGCGAGCGGGCGAGC
Consensus     (451)
                    501                                                550
NM_013476     (1)   ---------------------------------------------------
NM_000044     (501) TAGCTGCACATTGCAAAGAAGGCTCTTAGGAGCCAGGCGACTGGGGAGCG
Consensus     (501)
                    551                                                600
NM_013476     (1)   ---------------------------------------------------
NM_000044     (551) GCTTCAGCACTGCAGCCACGACCCGCCTGGTTAGGCTGCACGCGGAGAGA
Consensus     (551)
                    601                                                650
NM_013476     (1)   ---------------------------------------------------
NM_000044     (601) ACCCTCTGTTTTCCCCCACTCTCTCTCCACCTCCTCCTGCCTTCCCCACC
Consensus     (601)
                    651                                                700
NM_013476     (1)   ---------------------------------------------------
NM_000044     (651) CCGAGTGCGGAGCCAGAGATCAAAAGATGAAAAGGCAGTCAGGTCTTCAG
Consensus     (651)
                    701                                                750
NM_013476     (1)   ---------------------------------------------------
NM_000044     (701) TAGCCAAAAAACAAAACAAACAAAAACAAAAAAGCCGAAATAAAAGAAAA
Consensus     (701)
                    751                                                800
NM_013476     (1)   ---------------------------------------------------
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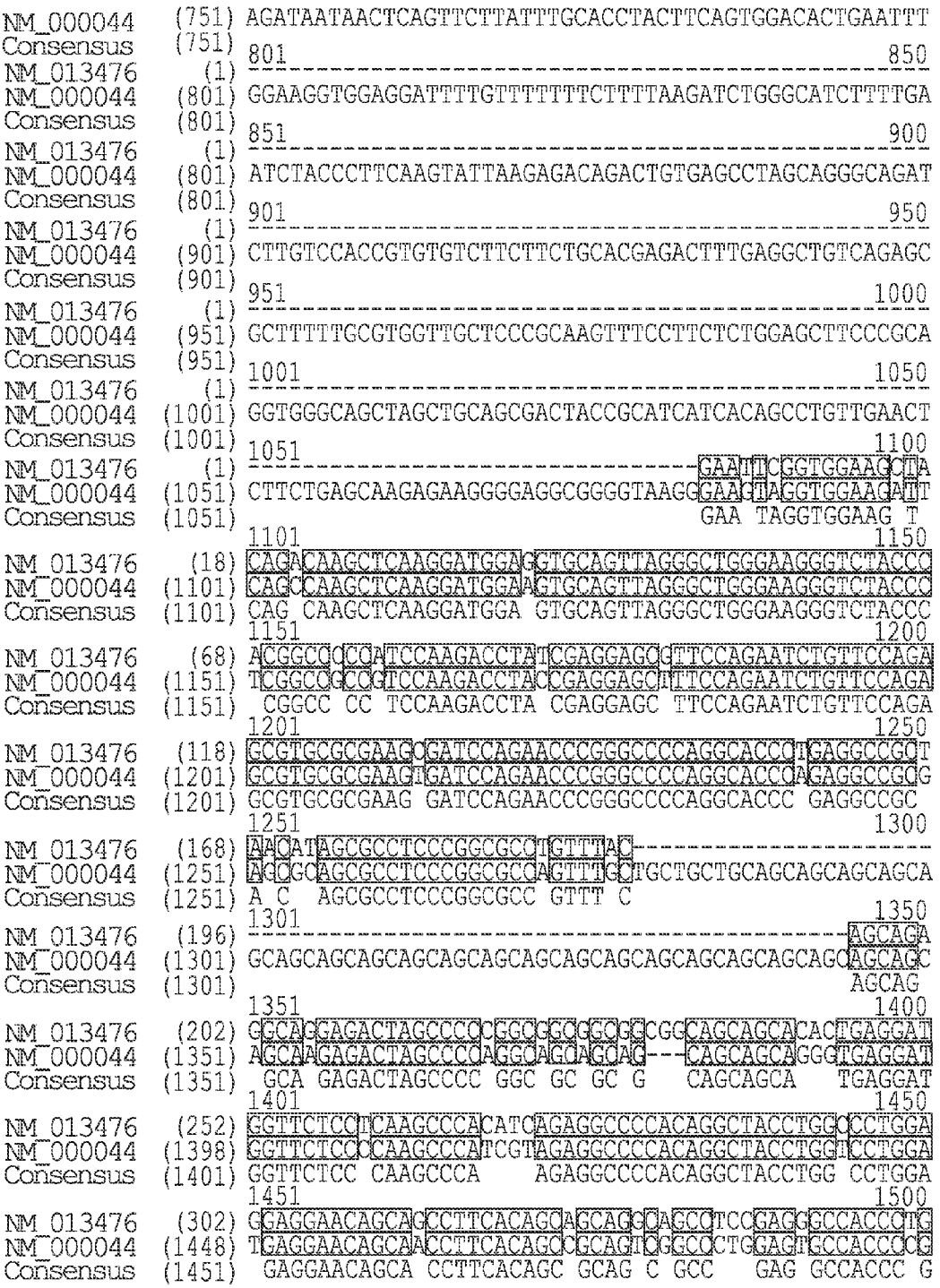
Figure 3 (cont'd)



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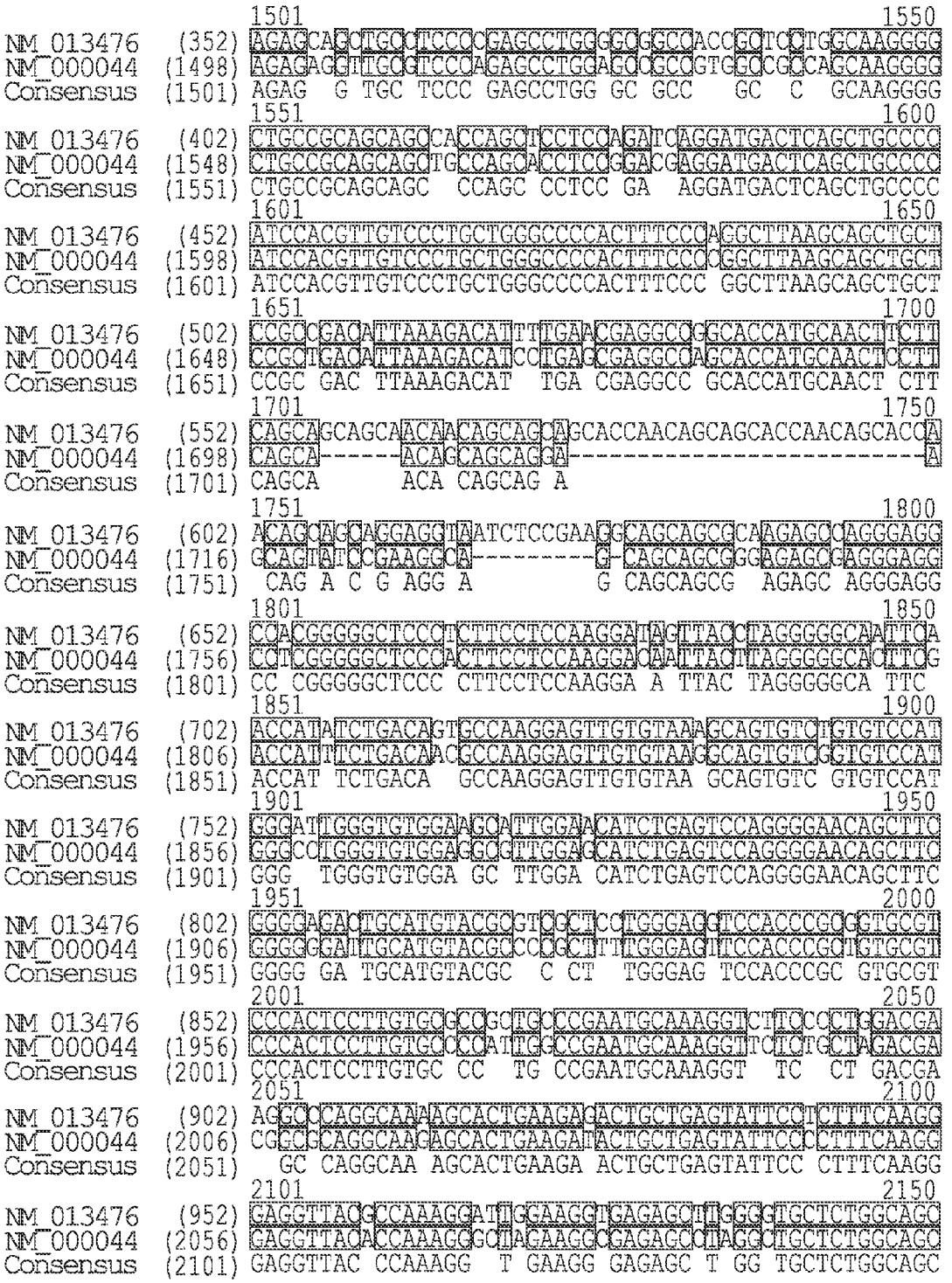

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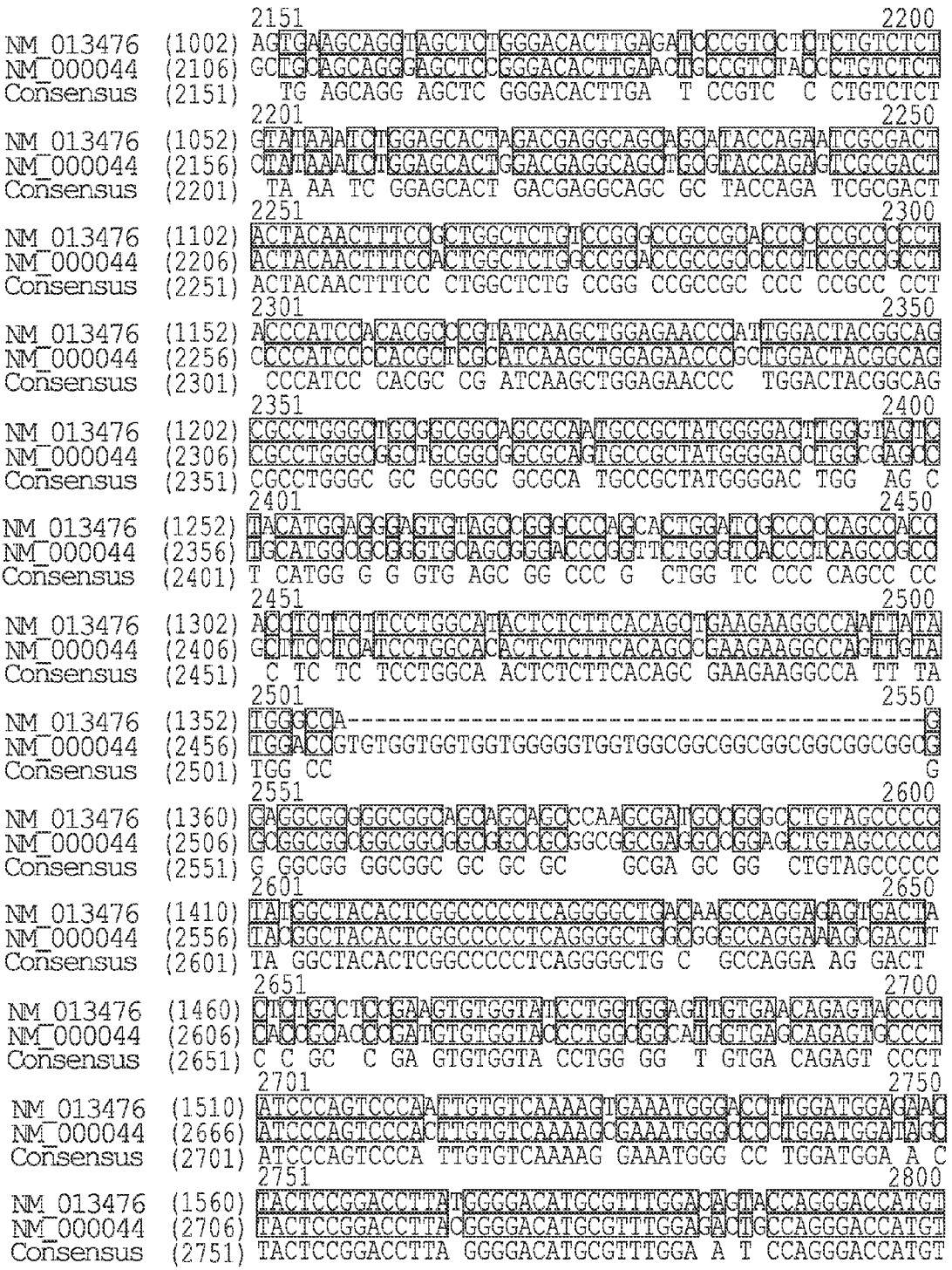

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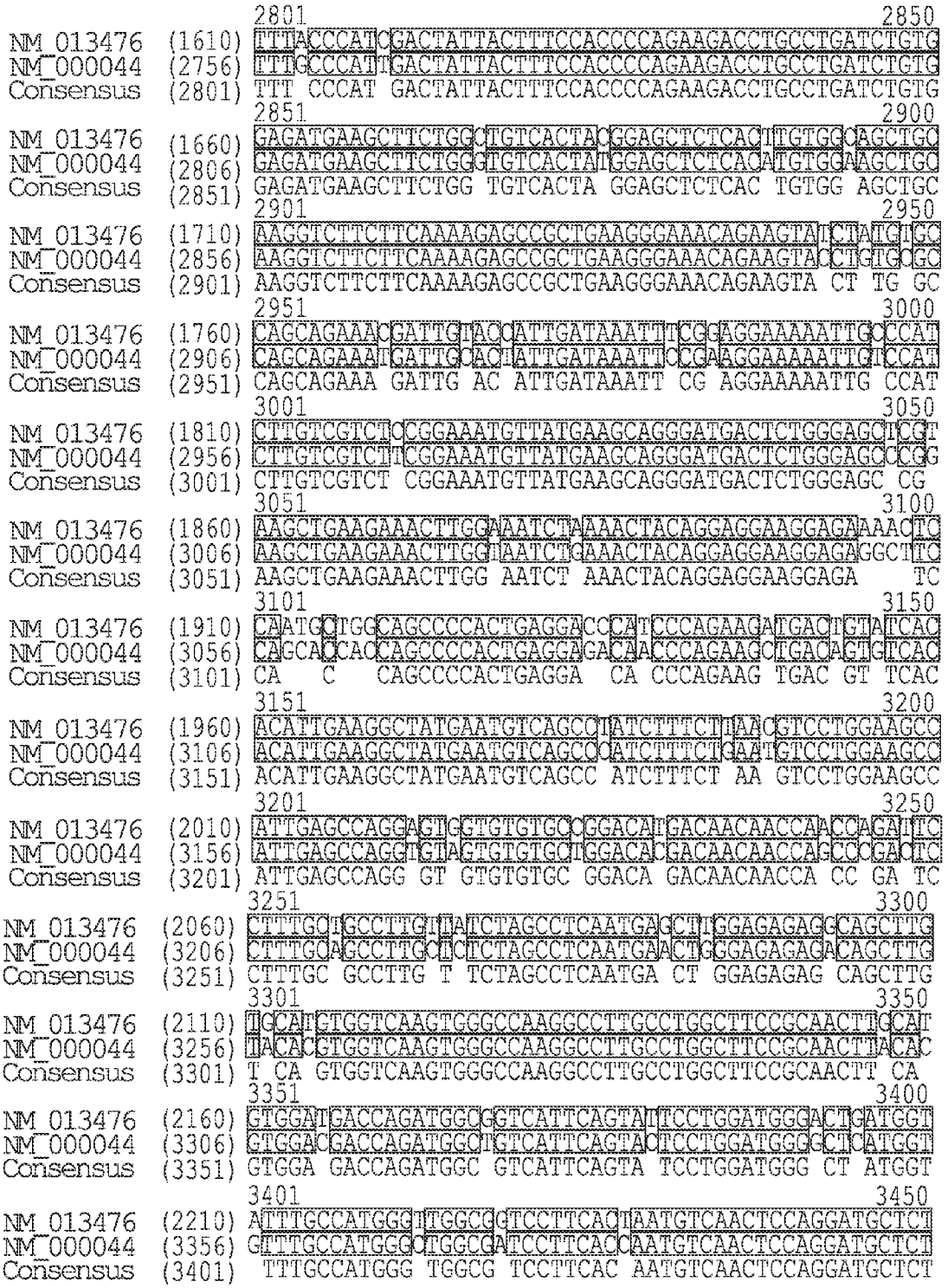

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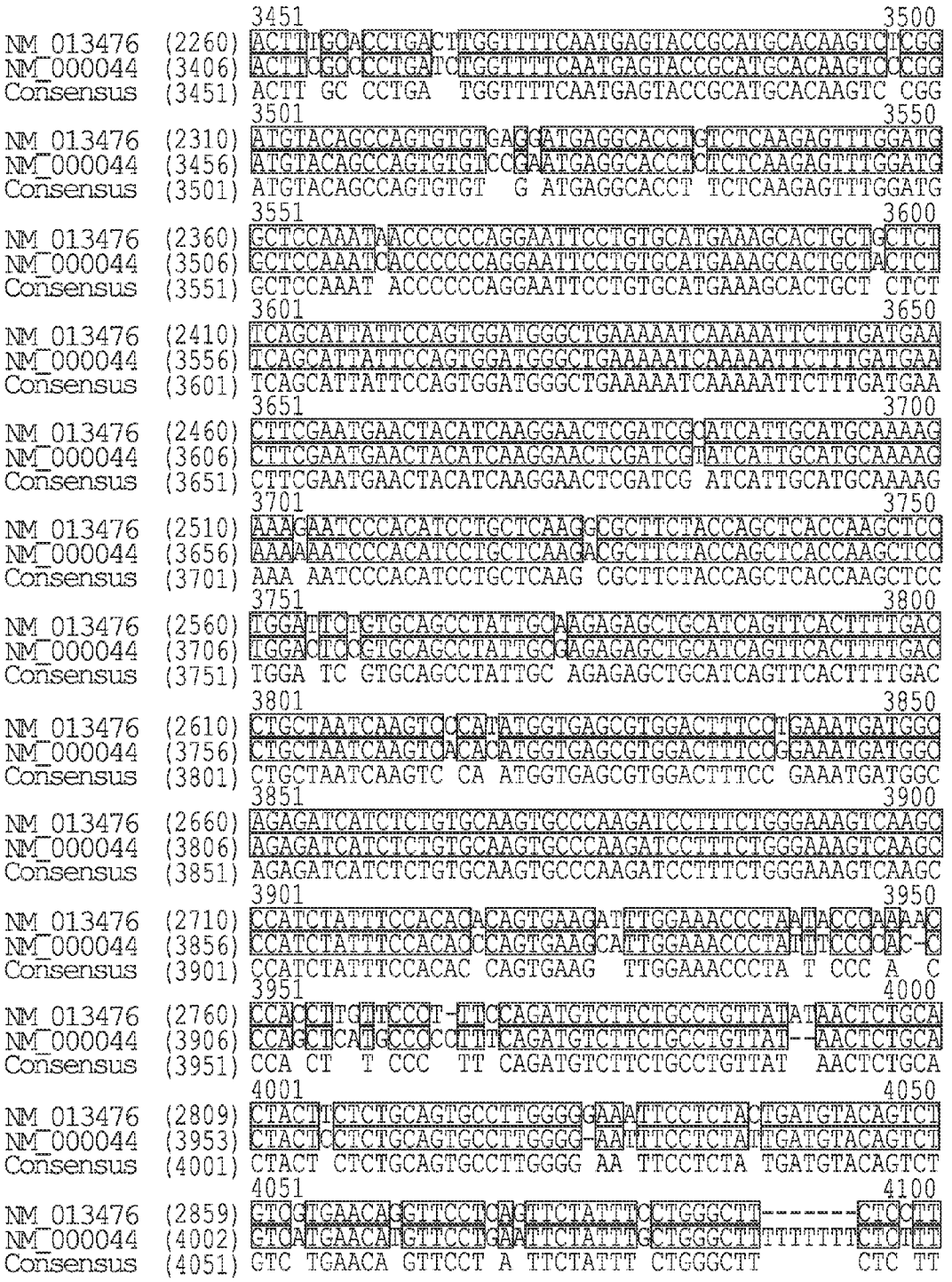

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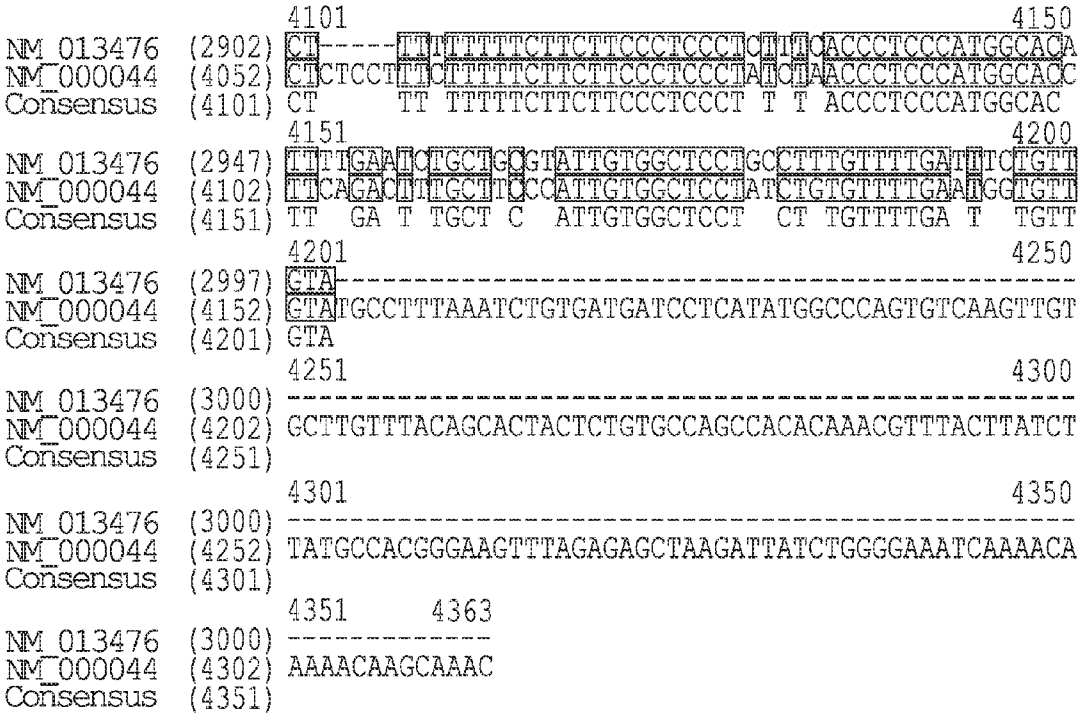

Figure 4

NM_000044

ORIGIN

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        1 cgagatcccg gggagccagc ttgctgggag agcgggacgg tccggagcaa gcccagaggc
       61 agaggaggcg acagagggaa aaagggccga gctagccgct ccagtgctgt acaggagccg
      121 aagggacgca ccacgccagc cccagcccgg ctccagcgac agccaacgcc tcttgcagcg
      181 cggcggcttc gaagccgccg cccggagctg ccctttcctc ttcggtgaag ttttaaaaag
      241 ctgctaaaga ctcggaggaa gcaaggaaag tgcctggtag gactgacggc tgcctttgtc
      301 ctcctcctct ccaccccgcc tccccccacc ctgccttccc ccctccccc gtcttctctc
      361 ccgcagctgc ctcagtcggc tactctcagc caaccccct caccaccctt ctccccaccc
      421 gccccccgc ccccgtggc ccagcgctgc cagccgagt ttgcagagag gtaactccct
      481 ttggctgcga gcgggcgagc tagctgcaca ttgcaaagaa ggctcttagg agccaggcga
      541 ctggggagcg gcttcagcac tgcagccacg acccgcctgg ttaggctgca cgcggagaga
      601 accctctgtt ttccccccact ctctctccac ctcctctgc cttccccacc ccgagtgccg
      661 agccagagat caaaagatga aaaggcagtc aggtcttcag tagccaaaaa acaaaacaaa
      721 caaaaacaaa aaagccgaaa taaaagaaaa agataataac tcagttctta tttgcaccta
      781 cttcagtgga cactgaattt ggaaggtgga ggattttgtt tttttctttt aagatctggg
      841 catcttttga atctaccctt caagtattaa gagacagact gtgagcctag cagggcagat
      901 cttgtccacc gtgtgtcttc ttctgcacga gactttgagg ctgtcagagc gcttttgcg
      961 tggttgctcc cgcaagtttc cttctctgga gcttcccgca ggtgggcagc tagctgcagc
     1021 gactaccgca tcatcacagc ctgttgaact cttctgagca agagaagggg aggcgggta
     1081 agggaagtag gtggaagatt cagccaagct caaggatgga agtgcagtta gggctgggaa
     1141 gggtctaccc tcggccgccg tccaagacct accgaggagc tttccagaat ctgttccaga
     1201 gcgtgcgcga agtgatccag aacccgggcc ccaggcaccc agaggccgcg agcgcagcac
     1261 ctcccggcgc cagtttgctg ctgctgcagc agcagcagca gcagcagcag cagcagcagc
     1321 agcagcagca gcagcagcag cagcagcagc agcaagagac tagccccagg cagcagcagc
```

SEQ ID NO:44

```
     1381 agcagcaggg tgaggatggt tctccccaag cccatcgtag aggcccacca ggctacctgg
     1441 tcctggatga ggaacagcaa cctcacagcc cgcagtcggc cctggagtgc cacccgaga
     1501 gaggttgcgt cccagagcct ggagccgccg tggccgccag caagggctg ccgcagcagc
     1561 tgccagcacc tcgggacgag gatgactcag ctgcccccatc cacgttgtcc ctgctgggcc
     1621 ccactttccc cggcttaagc agctgctccg ctgaccttaa agacatcctg agcgaggcca
     1681 gcaccatgca actccttcag caacagcagc aggaagcagt atccgaaggc agcagcagcg
     1741 ggagagcgag ggaggcctcg ggggctccca cttcctccaa ggacaattac ttagggggca
     1801 cttcgaccat ttctgacaac gccaaggagt tgtgtaaggc agtgtcggtg tccatgggcc
```

SEQ ID NO:45

SEQ ID NO:46

```
     1861 tgggtgtgga ggcgttggag catctgagtc cagggggaaca gcttcggggg gattgcatgt
```

SEQ ID NO:47

```
     1921 acgccccact tttgggagtt ccacccgctg tgcgtcccac tccttgtgcc ccatggccag
     1981 aatgcaaagg ttctctgcta gacgacagcg cagtcaagag cactgaagat actgctgagt
     2041 attcccctct caagggaggt tacaccaaag ggctagaagg cgagagccta ggctgctctg
     2101 gcagcgctgc agcagggagc tccgggacac ttgaactgcc gtctaccctg tctctctaca
     2161 agtccggagc actggacgag gcagctgcgt accagagtcg cgactactac aacttccac
     2221 tggctctggc cggaccgccg ccccctccgc cgcctccca tccccacgct cgcatcaagc
     2281 tggagaaccc gctggactac ggcagcgcct gggcggctgc ggcggcgcag tgccgctatg
     2341 ggacctggc gagcctgcat ggcgcgggtg cagcgggacc cggttctggg tcaccctcag
```

SEQ ID NO:48

```
     2401 ccgccgcttc ctcatcctgg cacactctct tcacagccga agaaggccag ttgtatggac
     2461 cgtgtggtgg tggtgggggt ggtggcggcg gcggcggcgg cggcggcggc ggcggcggcg
     2521 gcggcggcgg cggcgaggcg ggagctgtag ccccctacgg ctacactcgg ctccctcagg
     2581 ggctggcggg ccaggaaagc gacttcaccg cacctgatgt gtggtaccct ggcggcatgg
```

SEQ ID NO:51

```
     2641 tgagcagagt gccctatccc agtcccactt gtgtcaaaag cgaaatgggc ccctggatgg
     2701 atagctacta cggaccttac ggggacatgc gttcggagac tgccagggac catgttttgc
```

SEQ ID NO:54

```
     2761 ccattgacta ttactttcca ccccagaaga cctgcctgat ctgtggagat gaagcttctg
     2821 ggtgtcacta tggagctctc acatgtggaa gctgcaaggt cttctccaaa agagccgctg
     2881 aagggaaaca gaagtacctg tgcgccagca gaaatgattg cactattgat aaattccgaa
```

SEQ ID NO:57

```
     2941 ggaaaaattg tccatcttgt cgtcttcgga aatgttatga agcagggatg actctgggag
```

SEQ ID NO:58

Figure 4 (cont'd)

```
3001 cccggaaggct gaagaaactt ggtaatctga aactacagga ggaaggagag gcttccagca
3061 ccaccagccc cactgaggag acaacccaga agctgacagt gtcacacatt gaaggctatg
      SEQ ID NO:80
3121 aatgtcagcc catctttctg aatgtcctgg aagccattga gccaggtgta gtgtgtgctg
3181 gacacgacaa caaccagccc gactcctttg cagccttgct ctctagcctc aatgaactgg
                                    SEQ ID NO:61
3241 gagagagaca gcttgtacac gtggtcaagt gggccaaggc cttgcctggc ttccgcaact
3301 tacacgtgga cgaccagatg gctgtcattc agtactcctg gatggggctc atggtgtttg
                                    SEQ ID NO:62
3361 ccatgggctg gcgatccttc accaatgtcx actccaggat gctctacttc gcccctgatc
                                                SEQ ID NO:63
3421 tggttttcaa tgagtaccgc atgcacaagt cccggatgta cagccagtgt gtccgaatga
                    SEQ ID NO:66                                SEQ ID
NO:67
3481 ggcacctctc tcaagagttt ggatggctcc aaatcacccc ccaggaattc ctgtgcatga
                                                                SEQ ID
NO:68
3541 aagcactgct actcttcagc attattccag tggatgggct gaaaaatcaa aaattctttg
                                SEQ ID NO:69
3601 atgaacttcg aatgaactac atcaaggaac tcgatcgtat cattgcatgc aaaagaaaaa
3661 atcccacatc ctgctcaaga cgcttctacc agctcaccaa gctctggac tccgtgcagc
3721 ctattgcgag agagctgcat cagttcactt ttgacctgct aatcaagtca cacatggtga
                                        SEQ ID NO:72
3781 gcgtggactt tcccgaaatg atggcagaga tcatctctgt gcaagtgccc aagatccttt
          SEQ ID NO:73
3841 ctggaaaagt caagcccatc tatttccaca cccagtgaag cattggaaac cctatttccc
                                    SEQ ID NO:76
3901 caccccagct catgcccctt ttcagatgtc ttctgcctgt tataactctg cactactccc
          SEQ ID NO:77
3961 ctgcagtgcc ttggggaatt tcctctattg atgtacagtc tgtcatgaac atgttcctga
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4081 atctaacctt cccatggcac cttcagactt tgcttcccat tgtggctcct atctgtgttt
4141 tgaatggtgt tgtatgcctt taaatctgtg atgatcctca tatggcccag tgtcaagttg
4201 tgcttgttta cagcactact ctgtgccagc cacacaaacg tttacttatc ttatgccacg
4261 ggaagtttag agagctaaga ttatctgggg aaatcaaaac aaaaacaagc aaac
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Figure 5

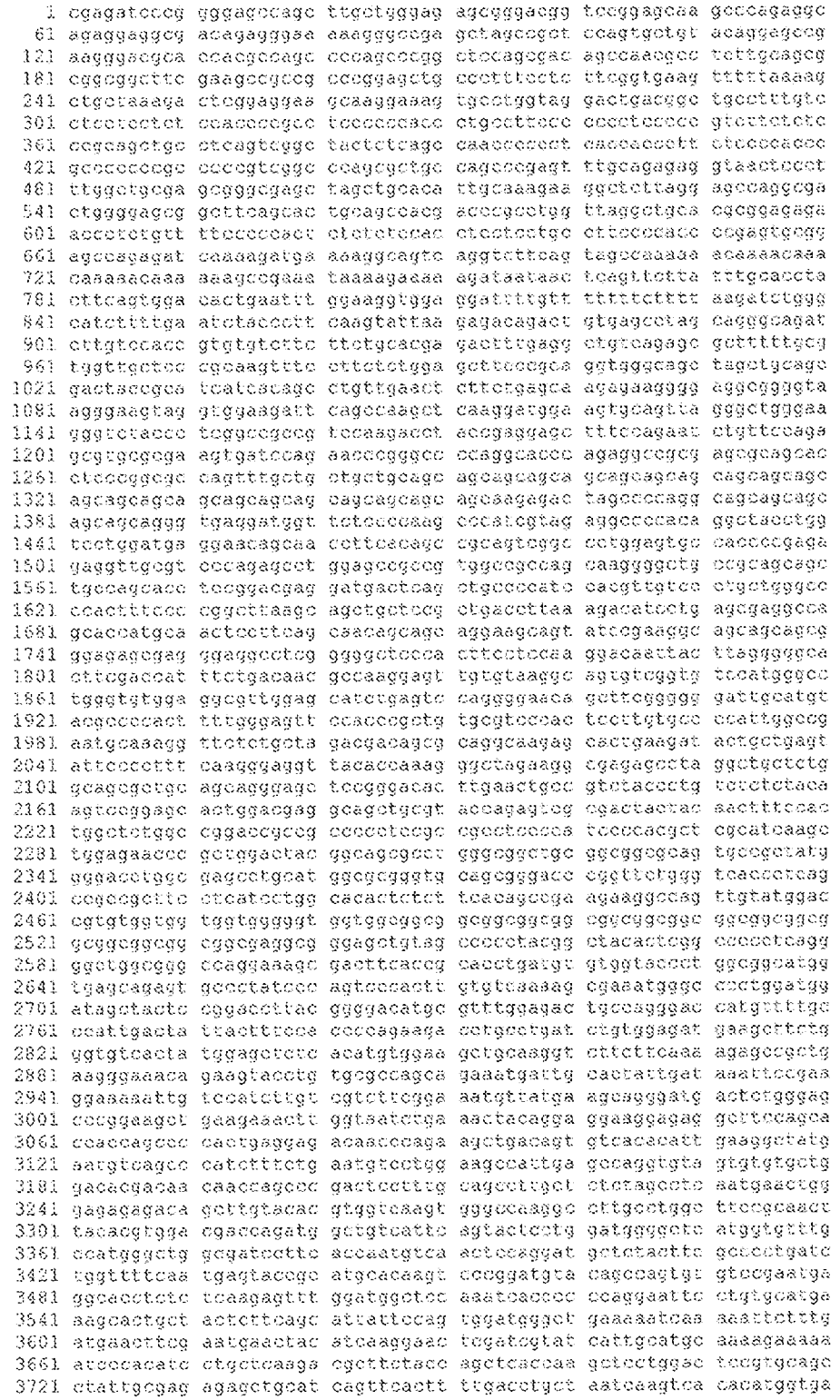

Figure 5 (cont'd)

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3781 gcgtggactt tccggaaatg atggcagaga tcatctctgt gcaagtgccc aagatccttt
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3901 cacccccagct catgcccctt ttcagatgtc ttctgcctgt tataactctg cactactcct
3961 ctgcagtgcc ttggggaatt tcctctattg atgtacagtc tgtcatgaac atgttcctga
4021 attctatttg ctgggctttt ttttttcttt tctttccttt cttttttctc ttccctcccc
4081 atctaaccct cccatggcac cttcagactt tgcttcccat tgtggctcct atctgtgttt
4141 tgaatggtgt tgtatgcctt taaatctgtg atgatcctca tatgcccaag tgtcaagttg
4201 tgcttgttta cagcactact ctgtgccagc cacacaaacg tttacttatc ttatgccacg
4261 ggaagtttag agagctaaga ttatctgggg aaatcaaaac aaaaacaagc aaac
//
```

Figure 6

SEQ ID NO: 81

```
LOCUS       NM_013476      2999 bp    mRNA    linear
DEFINITION  Mus musculus androgen receptor (Ar), mRNA.
ACCESSION   NM_013476
VERSION     NM_013476.3  GI:118129906
SOURCE      Mus musculus (house mouse)
ORIGIN
   1 gaattcggtg gaagctacag acaagctcaa ggatggaggt gcagttaggg ctgggaaggg
  61 tctacccacg gccccatcc aagacctatc gaggagcgtt ccagaatctg ttccagagcg
 121 tgcgcgaagc gatccagaac ccgggccccа ggcacctga ggccgctaac atagcacctc
 181 ccggcgcctg ttacagcag aggcaggaga ctagcccccg gcggcggcgg cggcagcagc
 241 acactgagga tggttctcct caagcccaca tcagaggccc cacaggctac ctggccctgg
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 421 ctcctccaga tcaggatgac tcagctgccc catccacgtt gtccctgctg ggccccactt
 481 tcccaggctt aagcagctgc tccgccgaca ttaaagacat tttgaacgag gccggcacca
 541 tgcaacttct tcagcagcag caacaacagc agcagcacca acagcagcac caacagcacc
 601 aacagcagca ggaggtaatc tccgaaggca gcagcgcaag agccagggag gccacggggg
 661 ctcctctttc ctccaaggat agttacctag ggggcaattc aaccatatct gacagtgcca
 721 aggagttgtg taaagcagtg tctgtgtcca tgggattggg tgtggaagca ttggaacatc
 781 tgagtccagg ggaacagctt cggggagact gcatgtacgc gtcgctcctg ggaggtccac
 841 ccgcggtgcg tcccactcct tgtgcgccgc tgcccgaatg caaaggtctt cccctggacg
 901 aaggcccagg caaaagcact gaagagactg ctgagtattc ctctttcaag ggaggttacg
 961 ccaaaggatt ggaaggtgag agcttggggt gctctggcag cagtgaagca ggtagctctg
1021 ggacacttga gatccgtcc tctctgtctc tgtataaatc tggagcacta gacgaggcag
1081 cagcatacca gaatcgcgac tactacaact ttccgctggc tctgtccggg ccgccgcacc
1141 ccccgccccc taccccatcca cacgcccgta tcaagctgga gcacccattg gactacggca
1201 gcgcctgggc tgcggcggca gcgcaatgcc gctatgggga cttgggtagt ctacatggag
1261 ggagtgtagc cgggcccagc actggatcgc ccccagccac cacctcttct tcctggcata
1321 ctctcttcac agctgaagaa ggccaattat atgggccagg aggcgggggc ggcagcagca
1381 gcccaagcga tgccgggcct gtagcccct atggctacac tcggccccct cagggctga
1441 caagccagga gagtgactac tctgcctccg aagtgtggta tcctggtgga gttgtgaaca
1501 gagtacccta tcccagtccc aattgtgtca aaagtgaaat gggaccttgg atggagaact
1561 actccggacc ttatggggac atgcgtttgg acagtaccag ggaccatgtt ttacccatcg
1621 actattactt tccacccccag aagacctgcc tgatctgtgg agatgaagct tctggctgtc
1681 actacggagc tctcacttgt ggcagctgca aggtcttctt caaaagagcc gctgaaggga
1741 aacagaagta tctatgtgcc agcagaaacg attgtaccat tgataaattt cggaggaaaa
1801 attgcccatc ttgtcgtctc cggaaatgtt atgaagcagg gatgactctg ggagctcgta
1861 agctgaagaa acttggaaat ctaaaactac aggaggaagg agaaaactcc aatgctggca
1921 gccccactga ggacccatcc cagaagatga ctgtatcaca cattgaaggc tatgaatgtc
1981 agcctatctt tcttaacgtc ctggaagcca ttgagccagg agtggtgtgt gccggacatg
2041 acaacaacca accagattcc tttgctgcct tgttatctag cctcaatgag cttggagaga
2101 ggcagcttgt gcatgtggtc aagtgggcca aggccttgcc tggcttccgc aacttgcatg
2161 tggatgacca gatggcggtc attcagtatt cctggatggg actgatggta tttgccatgg
2221 gttggcggtc cttcactaat gtcaactcca ggatgctcta ctttgcacct gacttggttt
2281 tcaatgagta ccgcatgcac aagtctcgga tgtacagcca gtgtgtgagg atgaggcacc
2341 tgtctcaaga gtttggatgg ctccaaataa ccccccagga attcctgtgc atgaaagcac
2401 tgctgctctt cagcattatt ccagtggatg ggctgaaaaa tcaaaaattc tttgatgaac
2461 ttcgaatgaa ctacatcaag gaactcgatc gcatcattgc atgcaaaaga aagaatccca
2521 catcctgctc aaggcgcttc taccagctca ccaagctcct ggattctgtg cagcctattg
2581 caagagagct gcatcagttc acttttgacc tgctaatcaa gtcccatatg gtgagcgtgg
2641 actttcctga aatgatggca gagatcatct ctgtgcaagt gcccaagatc ctttctggga
2701 aagtcaagcc catctatttc cacacacagt gaagatttgg aaaccctaat acccaaaacc
2761 cacctttgttc cctttccaga tgtcttctgc ctgttatata actctgcact actttctctgc
2821 agtgccttgg gggaaattcc tctactgatg tacagtctgt cgtgaacagg ttcctcagtt
2881 ctatttcctg ggcttctcct tcttttttt tcttcttccc tccctctttc acccttccat
2941 ggcacatttt gaatctgctg cgtattgtgg ctcctgcctt tgttttgatt tctgttgta
//
```

Figure 7

SEQ ID NO: 82

```
LOCUS       NM_001032911              3175 bp    mRNA
DEFINITION  Macaca mulatta androgen receptor (AR), mRNA.
ACCESSION   NM_001032911
VERSION     NM_001032911.1  GI:74136372
SOURCE      Macaca mulatta (rhesus monkey)

ORIGIN
        1 cccaaaaaat aaaaacaaac aaaaacaaaa caaaacaaaa aaaacgaata aagaaaaagg
       61 taataactca gttcttattt gcacctactt ccagtggaca ctgaatttgg aaggtggagg
      121 attcttgttt tttcttttaa gatcgggcat cttttgaatc tacrcctcaa gtgttaagag
      181 acagactgtg agcctagcag ggcagatctt gtccaccgtg tgtcttcttt tgcaggagac
      241 tttgaggctg tcagagcgct ttttgcgtgg ttgctcccgc aagtttcctt ctctggagct
      301 tcccgcaggt gggcagctag ctgcagcgac taccgcatca tcacagcctg ttgaactctt
      361 ctgagcaaga gaaggggagg cggggtaagg gaagtaggtg gaagattcag ccaagcttca
      421 ggatggaggt gcagttaggg ctggggaggg tctacctcg gccgccgtcc aagacctacc
      481 gaggagcttt ccagaatctg ttccagagcg tgcgcgaagt gatccagaac ccgggcccca
      541 ggcacccaga ggccgcgagc gcagcacctc ccggcgccag tttgcagcag cagcagcagc
      601 agcagcaaga aactagcccc cggcaacagc agcagcagca gcagggtgag gatggttctc
      661 cccaagccca tcgtagaggc cccacaggct acctggtcct ggatgaggaa cagcagcctt
      721 cacagcctca gtcagccccg gagtgccacc ccgagagagg ttgcgtccca gagcctggag
      781 ccgccgtggc cgccggcaag gggctgccgc agcagctgcc agcacctccg gacgaggatg
      841 actcagctgc cccatccacg ttgtctctgc tgggccccac tttccccggc ttaagcagct
      901 gctccgccga ccttaaagac atcctgagcg aggccagcac catgcaactc cttcagcaac
      961 agcagcagga agcagtatcc gaaggcagca gcagcgggag agcgagggag gcctcggggg
     1021 ctcccacttc ctccaaggac aattacttag agggcacttc gaccatttct gacagcgcca
     1081 aggagctgtg taaggcagtg tcggtgtcca tgggcttggg tgtggaggcg ttggagcatc
     1141 tgagtccagg ggaacagctt cggggggatt gcatgtacgc cccagttttg ggagttccac
     1201 ccgctgtgcg tcccactccg tgtgccccat tggccgaatg caaaggttct ctgctagacg
     1261 acagcgcagg caagagcact gaagatactg ctgagtattc ccctttcaag ggaggttaca
     1321 ccaaagggct agaaggcgag agcctaggct gctctggcag cgctgcagca gggagctccg
     1381 ggacacttga actgccgtcc accctgtctc tctacaagtc cggagcactg gacgaggcag
     1441 ctgcgtacca gagtcgcgac tactacaact ttccactggc tctggccggg ccgccgcccc
     1501 ctccaccgcc tccccatccc cacgctgcca tcaagctgga gaaccccgctg gactatggca
     1561 gcgcctgggc ggctgcggcg gcgcagtgcc gctatgggga cctggcgagc ctgcatggcg
     1621 cgggtgcagc gggacccggc tctgggtcac cctcagcggc cgcttcctca tcctggcaca
     1681 ctctcttcac agccgaagaa ggccagttgt atggaccgtg tggtggtggg ggcggcggcg
     1741 gtggcggcgg cggcggcggc gcaggcgagg cgggagctgt agccccctac ggctacactc
     1801 ggccacctca ggggctggcg ggccaggaag gcgacttcac cgcacctgat gtgtggtacc
     1861 ctggcggcat ggtgagcaga gtgccctatc ccagtcccac ttgtgtcaaa agcgagatgg
     1921 gcccctggat ggatagctac tccggacctt acgggggacat gcgtttggag actgccaggg
     1981 accatgtttt gccaattgac tattactttc cacccccagaa gaacctgcctg atctgtggag
     2041 atgaagcttc tgggtgtcac tatggagctc tcacatgtgg aagctgcaag gtcttcttca
     2101 aaagagccgc tgaagggaaa cagaagtacc tgtgtgccag cagaaatgat tgcactattg
     2161 ataaattccg aaggaaaaat tgtccatctt gccgcttcag gaaatgttat gaagcaggga
     2221 tgactctggg agcccggaag ctgaagaaac ttggtaatct gaaactacag gaggaaggag
     2281 aggcttccag caccaccagc cccactgagg agacagccca gaagctgaca gtgtcacaca
     2341 ttgaaggcta tgaatgtcag cccatctttc tgaatgtcct ggaggccatt gagccaggtg
     2401 tggtgtgtgc tggacatgac aacaaccagc cagactcctt cgcagccttg ctctctagcc
     2461 tcaatgaact gggagagaga cagcttgtac atgtggtcaa gtgggccaag gccttgcctg
     2521 gcttccgcaa cttacacgtg gacgaccaga tggctgtcat tcagtactcc tggatggggc
     2581 tcatggtgtt tgccatggcc tggcgatcct tcaccaatgt caactccagg atgctctact
     2641 ttgccccctga tctggtttic aatgagtacc gcatgcacaa atcccggatg tacagccagt
     2701 gtgtccgaat gaggcacctc tctcaagagt ttggatggct ccaaatcacc ccccaggaat
     2761 tctgtgcat gaaagcgctg ctactcttca gcattattcc agtggatggg ctgaaaaatc
     2821 aaaaattctt tgatgaactt cgaatgaact acatcaagga actcgatcgt atcattgcat
     2881 gcaaaagaaa aaatcccaca tctgctcaa ggcgtttcta ccagctcacc aagctcctgg
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     3001 cacacatggt gagcgtggac tttccggaaa tgatggcaga gatcatctct gtgcaagtgc
     3061 ccaagatcct ttctgggaaa gtcaagccca tctatttcca cacccagtga agcattggaa
     3121 atcctattt cctcaccccc gctcatgccc cctttcagat gtcttctgcc tgtta
//
```

Figure 8

SEQ ID NO:83

```
LOCUS       NP_000035                920 aa
DEFINITION  androgen receptor isoform 1 [Homo sapiens].
ACCESSION   NP_000035
VERSION     NP_000035.2  GI:21322252
DBSOURCE    REFSEQ: accession NM_000044.2
SOURCE      Homo sapiens (human)

ORIGIN
        1 mevqlglgrv yprppsktyr gafqnlfqsv reviqnpgpr hpeaasaapp gaslllqgq
       61 qgqqqqqqqq qqqqqqqqqq etsprqqqqq qgedgspqah rrgptgylvl deeqqpsqpq
      121 salechperg cvpepgaava askglpqqlp appdeddsaa pstlsllgpt fpglsscsad
      181 lkdilseast mqllqqqqqe avsegsssgr areasgapts skdnylggts tisdnakelc
      241 kavsvsmglg vealehlspg eqlrgdcmya pllgvppavr ptpcaplaec kgsllddsag
      301 kstedtaeys pfkggytkgl egeslgcsgs aaagssgtle lpstlslyks galdeaaayq
      361 srdyynfpla lagppppppp phpharikle npldygsawa aaaaqcrygd laslhgagaa
      421 gpgsgspsaa assswhtlft aceggqlygpc gggggggggg ggggggggqg gggeagavap
      481 ygytrppqgl agqesdftap dvwypggmvs rvpypsptcv ksemgpwmds ysgpygdmrl
      541 etardhvlpi dyyfppqktc licgdeasgc hygaltcgsc kvffkraaeg kqkylcasrn
      601 dctidkfrrk ncpscrlrkc yeagmtlgar klkklgnlkl qeegeasstt spteettqkl
      661 tvshiegyec qpiflnvlea iepgvvcagh dnnqpdsfaa llsslnelge rqlvhvvkwa
      721 kalpgfrnlh vddqmaviqy swmglmvfam gwrsftnvns rmlyfapdlv fneyrmhksr
      781 mysqcvrmrh lsqefgwlqi tpqeflcmka llifsiipvd glknqkffde lrmnyikeld
      841 riiackrknp tscsrrfyql tklldsvqpi arelhqftfd llikshmvsv dfpemmaeii
      901 svqvpkilsg kvkpiyfhtq
//
```

Figure 9

SEQ ID NO:84

```
LOCUS       NP_038504                899 aa
DEFINITION  androgen receptor [Mus musculus].
ACCESSION   NP_038504
VERSION     NP_038504.1  GI:7304901
DBSOURCE    REFSEQ: accession NM_013476.3
SOURCE      Mus musculus (house mouse)

ORIGIN
        1 mevqlglgrv yprppsktyr gafqnlfqsv reaiqnpgpr hpeaaniapp gaclqqrqet
       61 sprrrrrqqh tedgspqahi rgptgylale eeqqpsqqqa aseghpessc lpepgaatap
      121 gkglpqqppa ppdqddsaap stlsllgptf pglsscsadi kdilneagtm qllqqqqqqq
      181 qhqqqhqqhq gqqevisegs sarareatga pssskdsylg gnstisdsak eickavsvsm
      241 glgvealehl spgeqlrgdc myasllggpp avrptpcapl peckglplde gpgksteeta
      301 eyssfkggya kglegeslgc sgsseagssg tleipsslsl yksgaldeae ayqnrdyynf
      361 plalsgpphp pppthphari klenpldygs awaaaaaqcr ygdlgslhgg svaqpstqsp
      421 pattssswht lftaeeqqly gpgggggsss psdagpvapy gytrppqglt sqesdysase
      481 vwypggvvnr vpypspncvk semgpwmeny sgpygdmrld strdhvlpid yyfppqktcl
      541 icgdeasgch ygaltcgsck vffkraaegk qkylcasrnd ctidkfrrkn cpscrlrkcy
      601 eagmtlgark lkklgnlklq eegensnags ptedpsqkmt vshiegyecq pifinvleai
      661 epgvvcaghd nnqpdsfaal lsslnelger qlvhvvkwak alpgfrnlhv ddqmaviqys
      721 wmglmvfamg wrsftnvnsr mlyfapdlvf neyrmhksrm ysqcvrmrhl sqefgwlqit
      781 pqeflcmkal llfsiipvdg lknqkffdel rmnyikeldr iiackrknpt scsrrfyqlt
      841 klldsvqpia relhqfttfd likshmvsvd fpemmaeiis vqvpkilsgk vkpiyfhtq
//
```

Figure 10

SEQ ID NO:85

```
LOCUS       NP_001028083              895 aa
DEFINITION  androgen receptor [Macaca mulatta].
ACCESSION   NP_001028083
VERSION     NP_001028083.1  GI:74136373
DBSOURCE    REFSEQ: accession NM_001032911.1
SOURCE      Macaca mulatta (rhesus monkey)

ORIGIN
        1 mevqlglgrv yprppsktyr gafqnlfqsv revlqnpgpr hpeaasaapp qaslqqqqqq
       61 qqetspraqq qqqqgedgsp qahrrgptgy lvldeeqqps qpqsapechp ergcvpepga
      121 avaagkglpq qlpappdedd saapstlsll gptfpglssc sadlkdilse astmqllqqq
      181 qqeavsegss sgrareasga ptsskdnyle gtstisdsak elckavsvsm glgvealehl
      241 spgeqlrgdc myapvlgvpp avrptpcapl aeckgslldd sagkstedta eyspfkggyt
      301 klegeslgc sgsaaagssg tlelpstlsl yksgaldeaa ayqsrdyynf plalagpppp
      361 pppphphari klenpldygs awaaaaaqcr ygdlaslhga gaagpgsgsp saaassswht
      421 lftaeegqly gpcggggggg ggggggagea gavapygytr ppqglagqeg dftapdvwyp
      481 ggmvsrvpyp sptcvksemg pwmdsysgpy gdmrletard hvlpidyyfp pqktclicgd
      541 easgchygal tcgsckvffk raaegkqkyl casrndctid kfrrkncpsc rlrkcyeagm
      601 tlgarklkkl gnlklqeege assttsptee taqkltvshi egyecqpifl nvleaiepgv
      661 vcaghdnnqp dsfaallssl nelgerqlvh vvkwakalpg frnlhvddqm aviqyswmgl
      721 mvfamgwrsf tnvnsrmlyf apdlvfneyr mhksrmysqc vrmrhlsqef gwlqitpqef
      781 lcmkalllfs iipvdglknq kffdelrmny ikeldriiac krknptscsr rfyqltklld
      841 svqpiarelh qftfdlliks hmvsvdfpem maeiisvqvp kilsgkvkpi yfhtq
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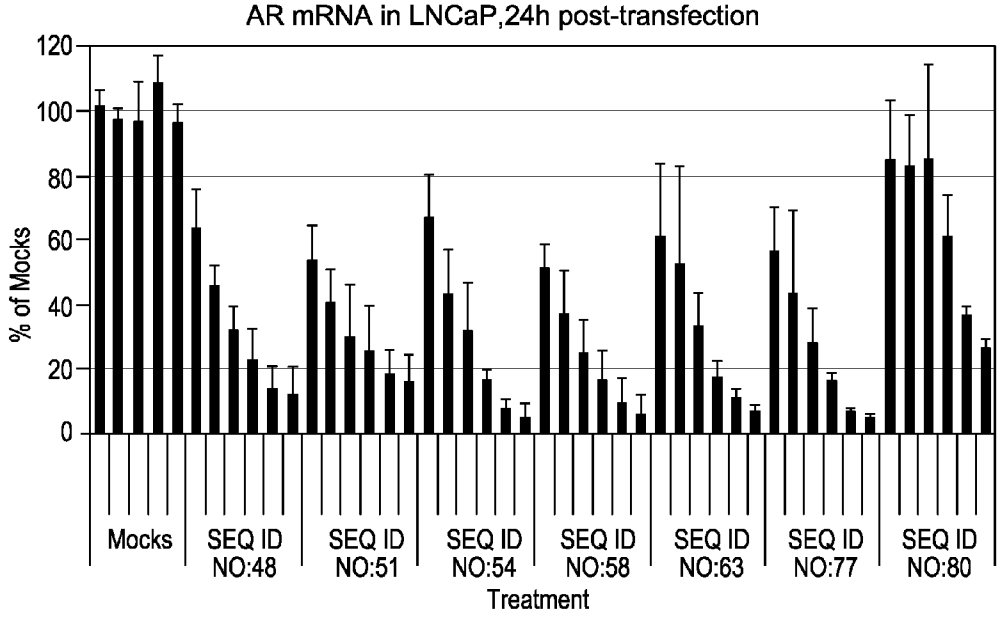


Figure 11

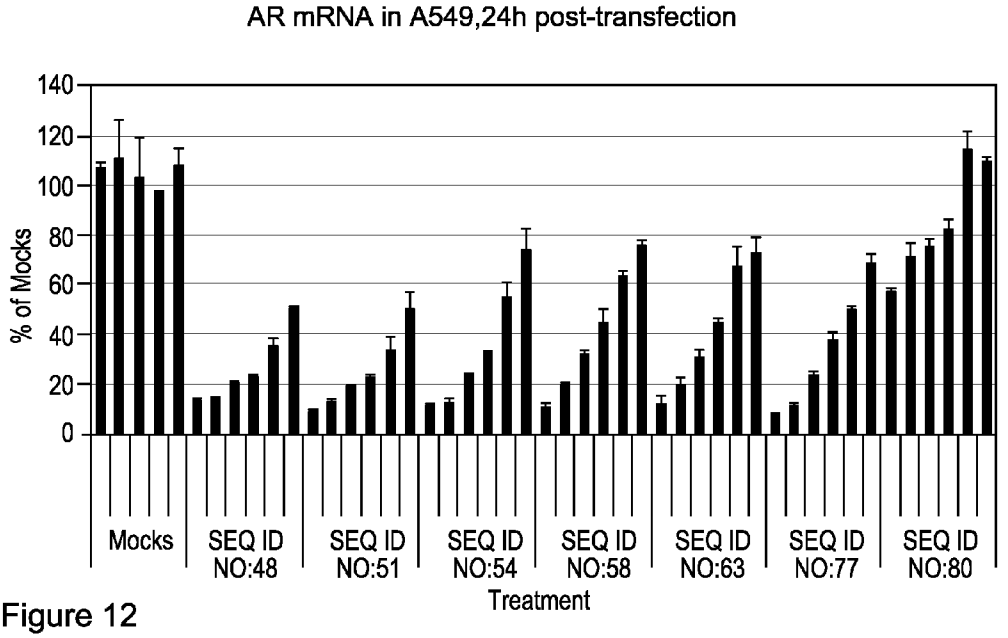
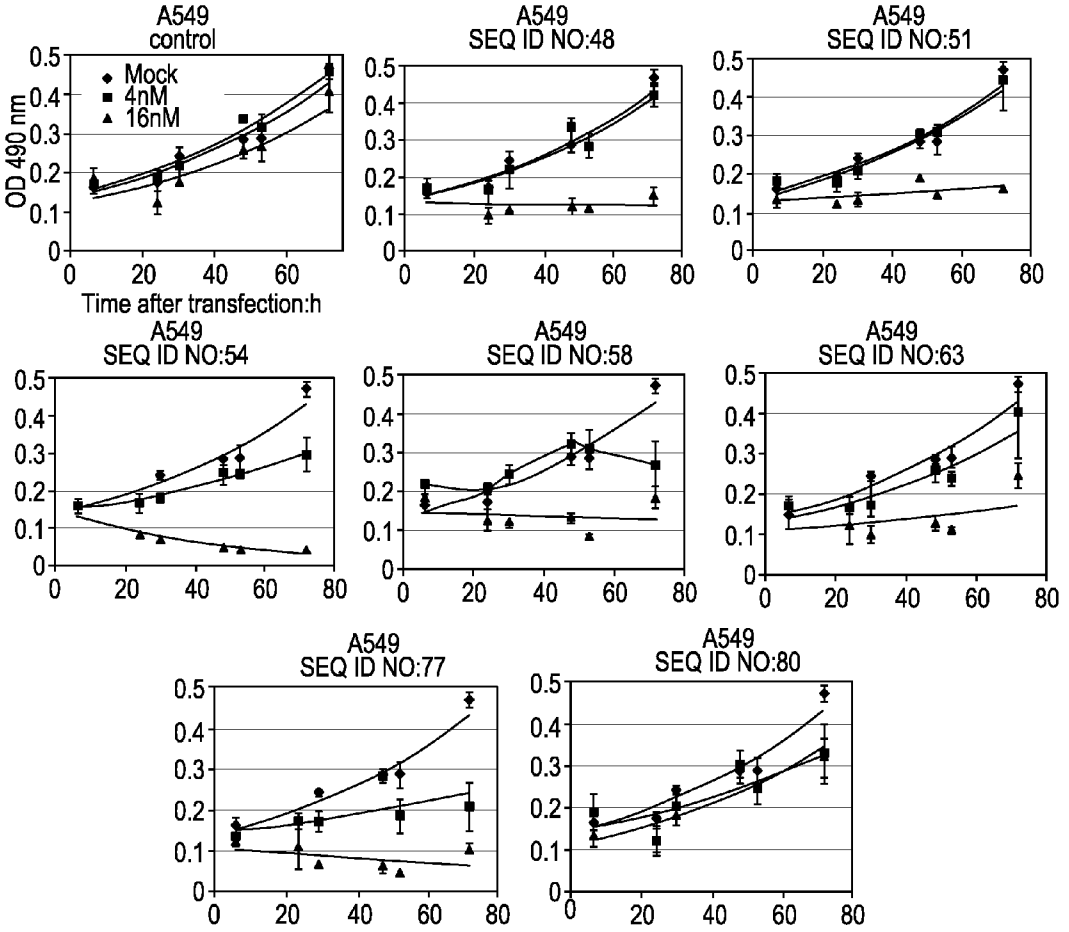


Figure 12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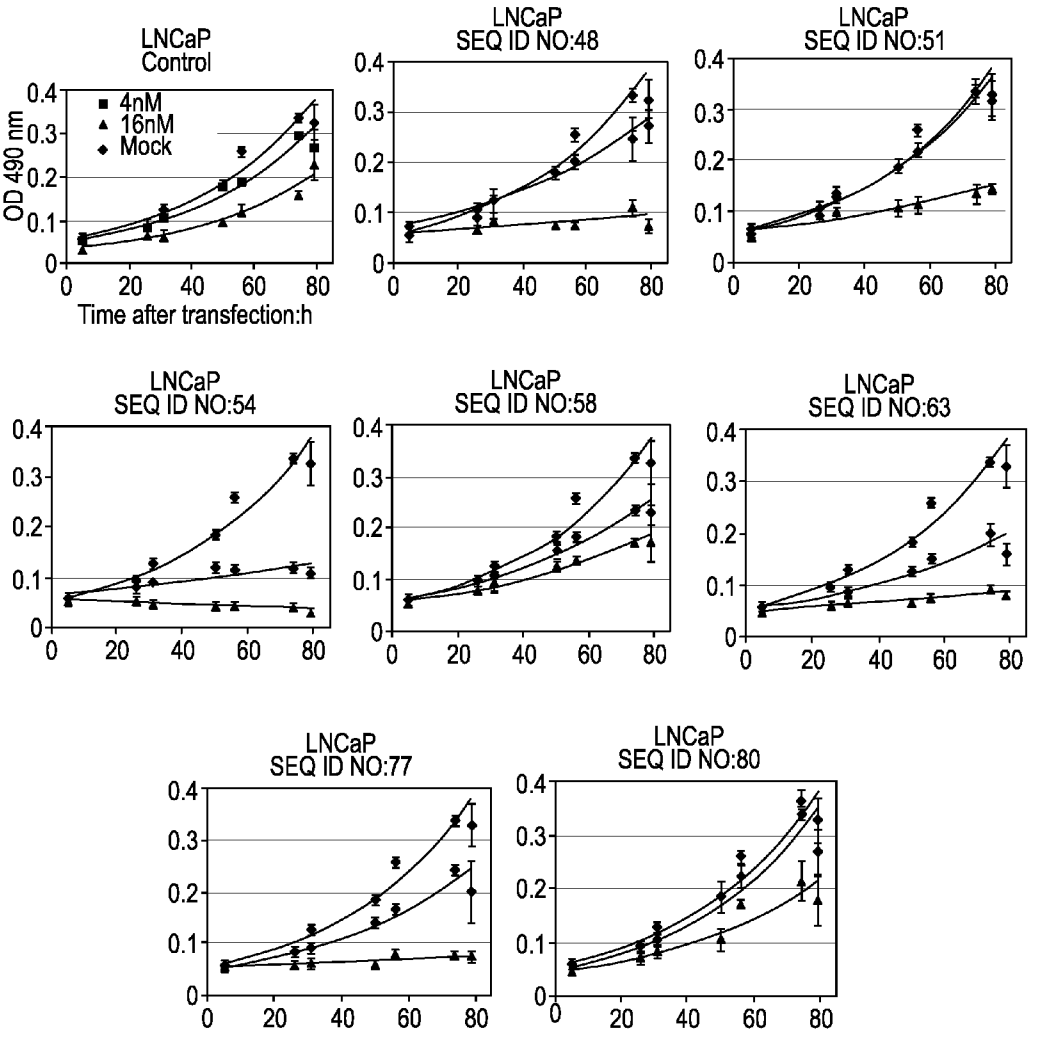
control
Mock
4nM
16nM
OD 490 nm
Time after transfection:h
A549
SEQ ID NO:48
A549
SEQ ID NO:51
A549
SEQ ID NO:54
A549
SEQ ID NO:58
A549
SEQ ID NO:63
A549
SEQ ID NO:77
A549
SEQ ID NO:80

Figure 13


LNCaP
Control
4nM
16nM
Mock
OD 490 nm
Time after transfection:h
LNCaP
SEQ ID NO:48
LNCaP
SEQ ID NO:51
LNCaP
SEQ ID NO:54
LNCaP
SEQ ID NO:58
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SEQ ID NO:63
LNCaP
SEQ ID NO:77
LNCaP
SEQ ID NO:80

Figure 14

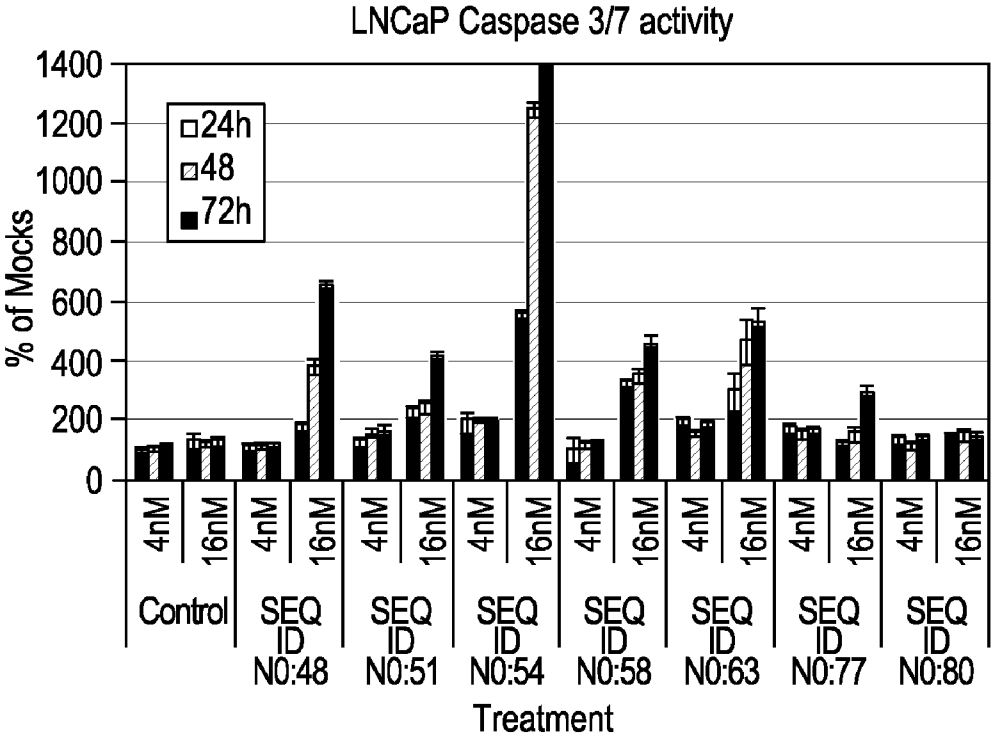


Figure 15

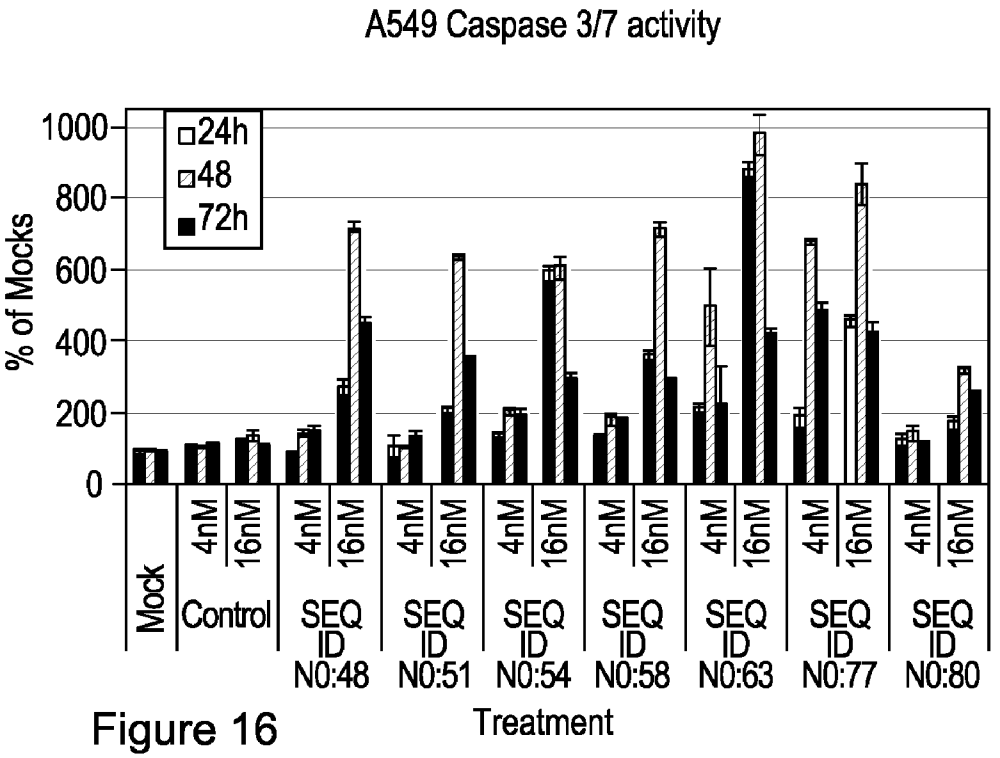


Figure 16

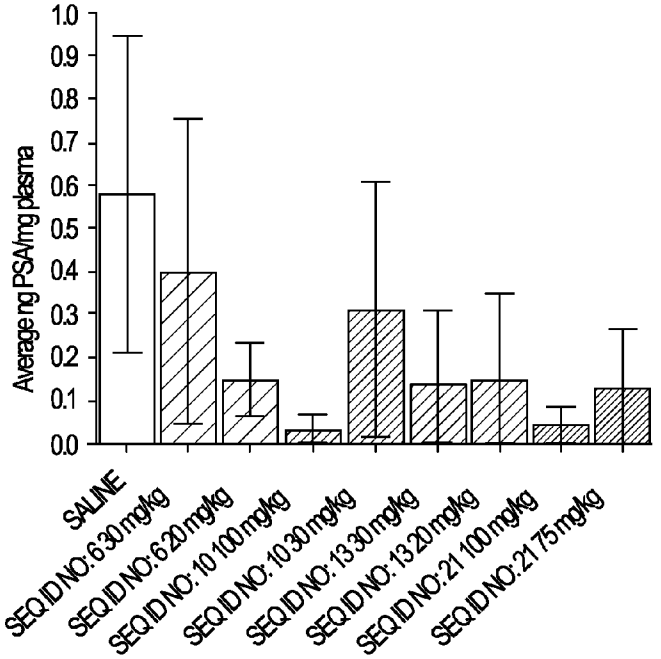


Figure 17

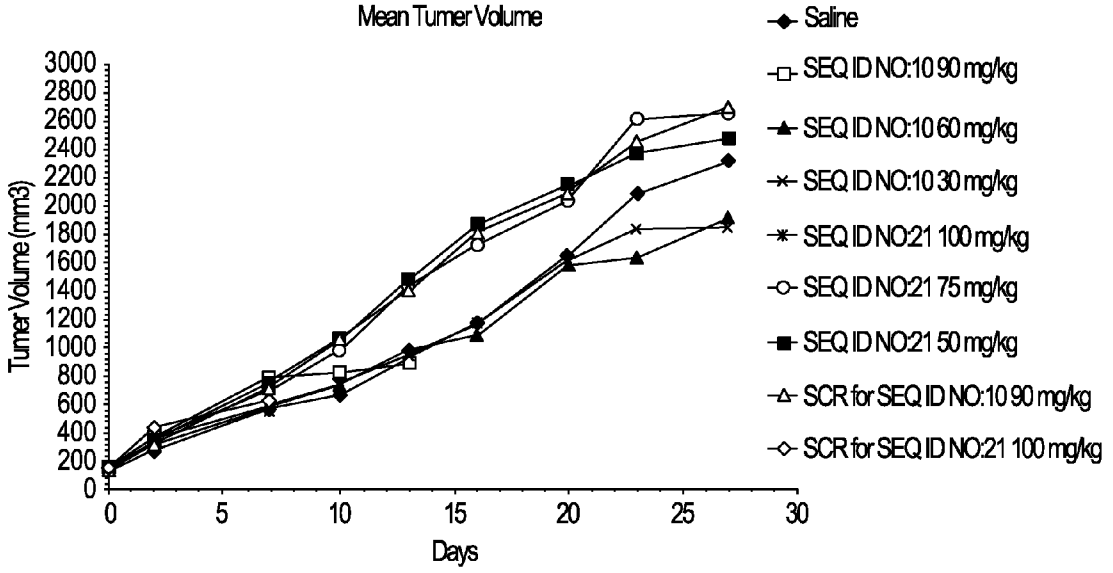


Figure 18

LNA ANTAGONISTS TARGETING THE ANDROGEN RECEPTOR

This application is a continuation application of U.S. application Ser. No. 12/322,033 filed on Nov. 26, 2008, which claims the benefit under 35 U.S.C. §119(e) of U.S. Provisional Application Ser. No. 60/990,125 filed Nov. 26, 2007, the disclosure of each of which is incorporated herein by reference in its entirety.

FIELD OF INVENTION

The invention provides compounds, compositions and methods for modulating the expression of the androgen receptor. In particular, this invention relates to oligomeric compounds (oligomers), which target the androgen receptor mRNA in a cell, leading to reduced expression of the androgen receptor. Reduction of androgen receptor expression is beneficial for a range of medical disorders, such as cancer, particularly prostate cancer or breast cancer.

BACKGROUND

The androgen receptor ("AR") is a type of nuclear receptor which is activated by binding of either of the androgenic hormones testosterone or dihydrotestosterone. The main function of the androgen receptor is as a DNA binding transcription factor which regulates gene expression. However the androgen receptor also has additional functions independent of DNA binding. The androgen receptor is most closely related to the progesterone receptor, and progestins in higher dosages can block the androgen receptor.

Whilst in humans the AR gene is single copy and found on the X chromosome at position Xq11-12, the receptor itself exists in two iso-forms (A and B). AR-A is an 87 kDa protein which lacks the first 187 amino acids (N-terminal truncation). Isoform AR-B is the full length 110 kDa version.

The binding of an androgen to the androgen receptor induces a conformational change in the receptor, resulting in a dissociation of heat shock proteins, dimerization and transport from the cytosol to the cell nucleus where the androgen receptor dimer binds to specific DNA sequences—referred to as hormone response elements. Depending on the interaction with other nuclear proteins, the AR controls gene expression, either increasing or decreasing transcription of specific genes, such as insulin-like growth factor I (IGF-1).

Androgen receptors can also have cytoplasmic activities through interaction with signal transduction proteins in the cytoplasm. Androgen binding to cytoplasmic androgen receptors can cause rapid changes in cell function independent of gene transcription, for example ion transport, as well as indirect influence of gene transcription, for example via mediating other signal transduction pathways, thereby influencing the activity of other transcription factors.

The over-expression of androgen receptor, or expression of mutated androgen receptor genes, has been indicated in several diseases, such as cancer, including prostate cancer and breast cancer, as well as other disorders such as polyglutamate disease (Monks et al., PNAS Nov. 2, 2007, published on line) alopecia, benign prostatic hyperplasia, spinal and muscular atrophy and Kennedy disease.

WO97/11170 describes a method of treating a patient diagnosed as having benign prostatic hyperplasia or a prostate cancer comprising administering an antisense oligonucleotide which selectively hybridises to the androgen receptor mRNA. Three antisense oligonucleotide sequences of between 27-29 nucleotides are disclosed.

U.S. Pat. No. 6,733,776 and EP 0 692 972 describe a method for treating androgenic alopecia by applying liposomes comprising an antisense nucleic acid that hybridises to an androgen receptor gene. No antisense molecules having specific sequences and targeting the androgen receptor are provided.

US 2005/0164970 describes a method of treating prostate cancer using siRNA complexes targeting the androgen receptor mRNA.

WO 2005/027833 describes a method of treating prostate cancer comprising administering to a patient an oligonucleotide comprising between 12-40 morpholino sub-units.

WO 2001/083740 describes an antisense compound having an uncharged morpholino backbone of between 18 to 20 contiguous units which targets the human androgen receptor.

Morpholino antisense compounds work via binding to the nucleic acid target to block access to the mRNA by other molecules, such as molecules involved in mRNA splicing or translation initiation.

U.S. Pat. No. 7,067,256 describes a ribozyme which apparently mediates inactivation of the androgen receptor. A 19-nucleotide RNA antisense molecule targeted to a region of the androgen receptor mRNA is provided.

However, despite the application of siRNA, morpholino-containing antisense oligonucleotides and ribozymes, none of the above androgen receptor inhibitors have been successful in efficiently down-regulating the androgen-receptor in vivo and at pharmacologically acceptable dosages.

The invention provides a new class of androgen receptor antagonists which contain locked nucleic acid ("LNA") monomers, and are targeted to particularly effective target sites on the androgen receptor mRNA.

SUMMARY OF INVENTION

The invention provides an oligomer of from 10-50 monomers, such as 10-30 monomers which comprises a first region of 10-50 monomers, such as 10-30 monomers, wherein the sequence of the first region is at least 80% (e.g., 85%, 90%, 95%, 98%, or 99%) identical to the reverse complement of a target region of a nucleic acid which encodes a mammalian androgen receptor, such as a mammalian androgen receptor gene or mRNA, such as a nucleic acid having the sequence set forth in SEQ ID NO: 1, or naturally occurring variants thereof. Thus, for example, the oligomer hybridizes to a region of a single-stranded nucleic acid molecule having the sequence shown in SEQ NO: 1.

The invention provides for a conjugate comprising the oligomer according to the invention, and at least one non-nucleotide or non-polynucleotide moiety covalently attached to the oligomer.

The invention provides for a pharmaceutical composition comprising the oligomer or the conjugate according to the invention, and a pharmaceutically acceptable diluent, carrier, salt or adjuvant.

The invention provides for the oligomer or the conjugate according to the invention, for use as a medicament, such as for the treatment of a disease or a medical disorder as disclosed herein, such as a hyperproliferative disorder, such as cancer or other hyperproliferative disorder. The invention provides for the use of an oligomer or the conjugate according to the invention, for the manufacture of a medicament for the treatment of a disease or disorder as disclosed herein, such as a hyperproliferative disorder, such as cancer.

The invention provides for a method of treating a disease or disorder as disclosed herein, such as a hyperproliferative disorder, such as cancer, the method comprising administer-

ing an oligomer, a conjugate or a pharmaceutical composition according to the invention to a patient suffering from or susceptible to the disease or disorder.

The invention provides for a method for the inhibition of androgen receptor in a cell which is expressing androgen receptor, the method comprising administering an oligomer, or a conjugate according to the invention to the cell so as to effect the inhibition of androgen receptor expression in said cell.

The invention provides an oligomer of from 10-50 monomers, which comprises a first region of 10-50 contiguous monomers, wherein the base sequence is at least 80% identical to the reverse complement of a target region of a nucleic acid which encodes a mammalian androgen receptor.

The invention further provides a conjugate comprising the oligomer according to the invention, which comprises at least one non-nucleotide or non-polynucleotide moiety ("conjugated moiety") covalently attached to the oligomer of the invention.

The invention provides for pharmaceutical compositions comprising an oligomer or conjugate of the invention, and a pharmaceutically acceptable diluent, carrier, salt or adjuvant.

The invention further provides for an oligomer according to the invention, for use in medicine.

The invention further provides for the use of the oligomer of the invention for the manufacture of a medicament for the treatment of one or more of the diseases referred to herein, such as a disease selected from the group consisting of cancer, such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease.

The invention further provides for an oligomer according to the invention, for use for the treatment of one or more of the diseases referred to herein, such as a disease selected from the group consisting of cancer, such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease.

Pharmaceutical and other compositions comprising an oligomer of the invention are also provided. Further provided are methods of down-regulating the expression of AR in cells or tissues comprising contacting said cells or tissues, in vitro or in vivo, with one or more of the oligomers, conjugates or compositions of the invention.

Also disclosed are methods of treating a non-human animal or a human suspected of having, or susceptible to, a disease or condition, associated with expression, or over-expression of AR by administering to the animal or human a therapeutically or prophylactically effective amount of one or more of the oligomers, conjugates or pharmaceutical compositions of the invention. Further, methods of using oligomers for the inhibition of expression of AR, and for treatment of diseases associated with activity of AR are provided.

The invention provides for a method for treating a disease selected from the group consisting of: cancer, such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease, the method comprising administering an effective amount of one or more oligomers, conjugates, or pharmaceutical compositions thereof to a patient in need thereof.

The invention provides for methods of inhibiting (e.g., by down-regulating) the expression of AR in a cell or a tissue, the method comprising the step of contacting the cell or tissue with an effective amount of one or more oligomers, conjugates, or pharmaceutical compositions thereof, to effect down-regulation of expression of AR.

BRIEF DESCRIPTION OF FIGURES

FIG. **1**. Oligonucleotides presented in Table 3 were evaluated for their potential to knockdown the androgen receptor mRNA at concentrations of 1, 4 and 16 nM in MCF7 cells 24 hours after transfection using Real-time PCR. All results were normalised to GAPDH and inhibition of AR mRNA is shown as percent of untreated control. Results shown are an average of three independent experiments.

FIG. **2**. Oligonucleotides presented in Table 3 were evaluated for their potential to knockdown the androgen receptor mRNA at concentrations of 1, 4 and 16 nM in A549 cells 24 hours after transfection using Real-time PCR. All results were normalised to GAPDH and inhibition of AR mRNA is shown as percent of untreated control. Results shown are an average of three independent experiments.

FIG. **3**. Sequence alignment of the human Androgen receptor mRNA sequence (GenBank Accession No.: NM_000044) and the mouse Androgen receptor mRNA sequence (GenBank Accession No.: NM_013476).

FIG. **4**. Location of presently preferred target regions of the human AR mRNA (cDNA) targeted by oligomers according to the invention. Although 16mer target sites have been shown, in some embodiments these target regions comprise an additional 4 monomers 5' or 3' to the target regions shown—i.e. are target regions comprising up to 24 contiguous monomers.

FIG. **5**. SEQ ID NO: 1 *Homo sapiens* androgen receptor (dihydrotestosterone receptor; testicular feminization; spinal and bulbar muscular atrophy; Kennedy disease) (AR), transcript variant 1, mRNA. (GenBank Accession number: NM_000044).

FIG. **6**. SEQ ID NO 81: Mouse androgen receptor mRNA sequence.

FIG. **7**. SEQ ID NO 82: Rhesus monkey androgen receptor mRNA sequence.

FIG. **8**. SEQ ID NO 83*: Homo sapiens* androgen receptor protein amino acid sequence.

FIG. **9**. SEQ ID NO 84: Mouse androgen receptor protein amino acid sequence.

FIG. **10**. SEQ ID NO 85: Rhesus monkey androgen receptor protein amino acid sequence.

FIG. **11**: AR mRNA in LNCaP, 24 h post-transfection

FIG. **12**: AR mRNA in A549, 24 h post-transfection

FIG. **13**: Cell proliferation assay—A549, time course post-transfection

FIG. **14**: Cell proliferation assay—time course post-transfection

FIG. **15**: Caspase 3/7 activity in LNCaP cells, 24, 48 or 72 hours post-transfection.

FIG. **16**: Caspase 3/7 activity in A549 cells, 24, 48 or 72 hours post-transfection.

FIG. **17**: Average PSA in plasma after in vivo oligomer treatment.

FIG. **18**: In vivo inhibition of tumor growth

DETAILED DESCRIPTION OF INVENTION

The Oligomer

The invention employs oligomeric compounds (referred herein as oligomers), for use in modulating the function of nucleic acid molecules encoding mammalian androgen receptor, such as the androgen receptor nucleic acid shown in SEQ ID NO: 1, and naturally occurring variants of such nucleic acid molecules encoding mammalian androgen receptor. The term "oligomer" in the context of the invention, refers to a molecule formed by covalent linkage of two or

more monomers (i.e. an oligonucleotide). In some embodiments, the oligomer comprises or consists of from 10-30 covalently linked monomers.

The term "monomer" includes both nucleosides and deoxynucleosides (collectively, "nucleosides") that occur naturally in nucleic acids and that do not contain either modified sugars or modified nucleobases, i.e., compounds in which a ribose sugar or deoxyribose sugar is covalently bonded to a naturally-occurring, unmodified nucleobase (base) moiety (i.e., the purine and pyrimidine heterocycles adenine, guanine, cytosine, thymine or uracil) and "nucleoside analogues," which are nucleosides that either do occur naturally in nucleic acids or do not occur naturally in nucleic acids, wherein either the sugar moiety is other than a ribose or a deoxyribose sugar (such as bicyclic sugars or 2' modified sugars, such as 2' substituted sugars), or the base moiety is modified (e.g., 5-methylcytosine), or both.

An "RNA monomer" is a nucleoside containing a ribose sugar and an unmodified nucleobase.

A "DNA monomer" is a nucleoside containing a deoxyribose sugar and an unmodified nucleobase.

A "Locked Nucleic Acid monomer," "locked monomer," or "LNA monomer" is a nucleoside analogue having a bicyclic sugar, as further described herein below.

The terms "corresponding nucleoside analogue" and "corresponding nucleoside" indicate that the base moiety in the nucleoside analogue and the base moiety in the nucleoside are identical. For example, when the "nucleoside" contains a 2-deoxyribose sugar linked to an adenine, the "corresponding nucleoside analogue" contains, for example, a modified sugar linked to an adenine base moiety.

The terms "oligomer," "oligomeric compound," and "oligonucleotide" are used interchangeably in the context of the invention, and refer to a molecule formed by covalent linkage of two or more contiguous monomers by, for example, a phosphate group (forming a phosphodiester linkage between nucleosides) or a phosphorothioate group (forming a phosphorothioate linkage between nucleosides). The oligomer consists of, or comprises, 10-50 monomers, such as 10-30 monomers.

In some embodiments, an oligomer comprises nucleosides, or nucleoside analogues, or mixtures thereof as referred to herein. An "LNA oligomer" or "LNA oligonucleotide" refers to an oligonucleotide containing one or more LNA monomers.

Nucleoside analogues that are optionally included within oligomers may function similarly to corresponding nucleosides, or may have specific improved functions. Oligomers wherein some or all of the monomers are nucleoside analogues are often preferred over native forms because of several desirable properties of such oligomers, such as the ability to penetrate a cell membrane, good resistance to extra- and/or intracellular nucleases and high affinity and specificity for the nucleic acid target. LNA monomers are particularly preferred, for example, for conferring several of the above-mentioned properties.

In various embodiments, one or more nucleoside analogues present within the oligomer are "silent" or "equivalent" in function to the corresponding natural nucleoside, i.e., have no functional effect on the way the oligomer functions to inhibit target gene expression. Such "equivalent" nucleoside analogues are nevertheless useful if, for example, they are easier or cheaper to manufacture, or are more stable under storage or manufacturing conditions, or can incorporate a tag or label. Typically, however, the analogues will have a functional effect on the way in which the oligomer functions to inhibit expression; for example, by producing increased binding affinity to the target region of the target nucleic acid and/or increased resistance to intracellular nucleases and/or increased ease of transport into the cell.

Thus, in various embodiments, oligomers according to the invention comprise nucleoside monomers and at least one nucleoside analogue monomer, such as an LNA monomer, or other nucleoside analogue monomers.

The term "at least one" comprises the integers larger than or equal to 1, such as 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20 and so forth. In various embodiments, such as when referring to the nucleic acid or protein targets of the compounds of the invention, the term "at least one" includes the terms "at least two" and "at least three" and "at least four." Likewise, in some embodiments, the term "at least two" comprises the terms "at least three" and "at least four."

In some embodiments, the oligomer comprises or consists of 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29 or 30 contiguous monomers.

In some embodiments, the oligomer comprises or consists of 10-22 contiguous monomers, such as 12-18 contiguous monomers, such as 13-17 or 12-16 contiguous monomers, such as 13, 14, 15, 16 contiguous monomers.

In certain embodiments, the oligomer comprises or consists of 10, 11, 12, 13, or 14 contiguous monomers.

In various embodiments, the oligomer according to the invention consists of no more than 22 monomers, such as no more than 20 monomers, such as no more than 18 monomers, such as 15, 16 or 17 monomers. In some embodiments, the oligomer of the invention comprises less than 20 monomers.

In various embodiments, the compounds of the invention do not comprise RNA monomers.

In various embodiments, the compounds according to the invention are linear molecules or are linear as synthesised. The oligomer, in such embodiments, is a single stranded molecule, and typically does not comprise short regions of, for example, at least 3, 4 or 5 contiguous monomers, which are complementary to another region within the same oligomer such that the oligomer forms an internal duplex. In some embodiments, the oligomer is essentially not double stranded, i.e., is not a siRNA.

In some embodiments, the oligomer of the invention consists of a contiguous stretch of monomers, the sequence of which is identified by a SEQ ID NO disclosed herein (see, e.g., Tables 1-3). In other embodiments, the oligomer comprises a first region, the region consisting of a contiguous stretch of monomers, and one or more additional regions which consist of at least one additional monomer. In some embodiments, the sequence of the first region is identified by a SEQ ID NO disclosed herein.

Gapmer Design

Typically, the oligomer of the invention is a gapmer.

A "gapmer" is an oligomer which comprises a contiguous stretch of monomers capable of recruiting an RNAse (e.g., such as RNAseH) as further described herein below, such as a region of at least 6 or 7 DNA monomers, referred to herein as region B, wherein region B is flanked both on its 5' and 3' ends by regions respectively referred to as regions A and C, each of regions A and C comprising or consisting of nucleoside analogues, such as affinity-enhancing nucleoside analogues, such as 1-6 nucleoside analogues.

Typically, the gapmer comprises regions, from 5' to 3', A-B-C, or optionally A-B-C-D or D-A-B-C, wherein: region A consists of or comprises at least one nucleoside analogue, such as at least one LNA monomer, such as 1-6 nucleoside analogues, such as LNA monomers, and region B consists of or comprises at least five contiguous monomers which are capable of recruiting RNAse (when formed in a duplex with

a complementary target region of the target RNA molecule, such as the mRNA target), such as DNA monomers; region C consists of or comprises at least one nucleoside analogue, such as at least one LNA monomer, such as 1-6 nucleoside analogues, such as LNA monomers; and region D, when present, consists of or comprises 1, 2 or 3 monomers, such as DNA monomers.

In various embodiments, region A consists of 1, 2, 3, 4, 5 or 6 nucleoside analogues, such as LNA monomers, such as 2-5 nucleoside analogues, such as 2-5 LNA monomers, such as 3 or 4 nucleoside analogues, such as 3 or 4 LNA monomers; and/or region C consists of 1, 2, 3, 4, 5 or 6 nucleoside analogues, such as LNA monomers, such as 2-5 nucleoside analogues, such as 2-5 LNA monomers, such as 3 or 4 nucleoside analogues, such as 3 or 4 LNA monomers.

In certain embodiments, region B consists of or comprises 5, 6, 7, 8, 9, 10, 11 or 12 contiguous monomers which are capable of recruiting RNAse, or 6-10, or 7-9, such as 8 contiguous monomers which are capable of recruiting RNAse. In certain embodiments, region B consists of or comprises at least one DNA monomer, such as 1-12 DNA monomers, preferably 4-12 DNA monomers, more preferably 6-10 DNA monomers, such as 7-10 DNA monomers, most preferably 8, 9 or 10 DNA monomers.

In various embodiments, region A consists of 3 or 4 nucleoside analogues, such as LNA monomers, region B consists of 7, 8, 9 or 10 DNA monomers, and region C consists of 3 or 4 nucleoside analogues, such as LNA monomers. Such designs include (A-B-C) 3-10-3, 3-10-4, 4-10-3, 3-9-3, 3-9-4, 4-9-3, 3-8-3, 3-8-4, 4-8-3, 3-7-3, 3-7-4, 4-7-3, and may further include region D, which may have one or 2 monomers, such as DNA monomers.

Further gapmer designs are disclosed in WO2004/046160, which is hereby incorporated by reference.

US provisional application, 60/977,409, hereby incorporated by reference, refers to 'shortmer' gapmer oligomers. In some embodiments, oligomers presented here may be such shortmer gapmers.

In certain embodiments, the oligomer consists of 10, 11, 12, 13 or 14 contiguous monomers, wherein the regions of the oligomer have the pattern (5'-3'), A-B-C, or optionally A-B-C-D or D-A-B-C, wherein: region A consists of 1, 2 or 3 nucleoside analogue monomers, such as LNA monomers; region B consists of 7, 8 or 9 contiguous monomers which are capable of recruiting RNAse when formed in a duplex with a complementary RNA molecule (such as a mRNA target); and region C consists of 1, 2 or 3 nucleoside analogue monomers, such as LNA monomers. When present, region D consists of a single DNA monomer.

In certain embodiments, region A consists of 1 LNA monomer. In certain embodiments, region A consists of 2 LNA monomers. In certain embodiments, region A consists of 3 LNA monomers. In certain embodiments, region C consists of 1 LNA monomer. In certain embodiments, region C consists of 2 LNA monomers. In certain embodiments, region C consists of 3 LNA monomers. In certain embodiments, region B consists of 7 nucleoside monomers, In certain embodiments, region B consists of 8 nucleoside monomers. In certain embodiments, region B consists of 9 nucleoside monomers. In certain embodiments, region B comprises 1-9 DNA monomers, such as 2, 3, 4, 5, 6, 7 or 8 DNA monomers. In certain embodiments, region B consists of DNA monomers. In certain embodiments, region B comprises at least one LNA monomer which is in the alpha-L configuration, such as 2, 3, 4, 5, 6, 7, 8 or 9 LNA monomers in the alpha-L-configuration. In certain embodiments, region B comprises at least one alpha-L-oxy LNA monomer. In certain embodiments, all the LNA monomers in region B that are in the alpha-L-configuration are alpha-L-oxy LNA units. In certain embodiments, the number of monomers present in the A-B-C regions are selected from the group consisting of (nucleoside analogue monomers—region B—nucleoside analogue monomers): 1-8-1, 1-8-2, 2-8-1, 2-8-2, 3-8-3, 2-8-3, 3-8-2, 4-8-1, 4-8-2, 1-8-4, 2-8-4, or; 1-9-1, 1-9-2, 2-9-1, 2-9-2, 2-9-3, 3-9-2, 1-9-3, 3-9-1, 4-9-1, 1-9-4, or; 1-10-1, 1-10-2, 2-10-1, 2-10-2, 1-10-3, 3-10-1. In certain embodiments, the number of monomers present in the A-B-C regions of the oligomer of the invention is selected from the group consisting of: 2-7-1, 1-7-2, 2-7-2, 3-7-3, 2-7-3, 3-7-2, 3-7-4, and 4-7-3. In certain embodiments, each of regions A and C consists of two LNA monomers, and region B consists of 8 or 9 nucleoside monomers, preferably DNA monomers.

In various embodiments, other gapmer designs include those where regions A and/or C consists of 3, 4, 5 or 6 nucleoside analogue, such as monomers containing a 2'-O-methoxyethyl-ribose sugar (2'-MOE) or monomers containing a 2'-fluoro-deoxyribose sugar, and region B consists of 8, 9, 10, 11 or 12 nucleosides, such as DNA monomers, where regions A-B-C have 5-10-5 or 4-12-4 monomers. Further gapmer designs are disclosed in WO 2007/146511A2, hereby incorporated by reference.

Internucleoside Linkages

The monomers of the oligomers described herein are coupled together via linkage groups. Suitably, each monomer is linked to the 3' adjacent monomer via a linkage group.

The terms "linkage group" or "internucleoside linkage" means a group capable of covalently coupling together two contiguous monomers. Specific and preferred examples include phosphate groups (forming a phosphodiester between adjacent nucleoside monomers) and phosphorothioate groups (forming a phosphorothioate linkage between adjacent nucleoside monomers).

Suitable linkage groups include those listed in PCT/DK2006/000512, for example in the first paragraph of page 34 of PCT/DK2006/000512 (hereby incorporated by reference).

It is, in various embodiments, preferred to modify the linkage group from its normal phosphodiester to one that is more resistant to nuclease attack, such as phosphorothioate or boranophosphate—these two being cleavable by RNase H, thereby permitting RNase-mediated antisense inhibition of expression of the target gene.

In some embodiments, suitable sulphur (S) containing linkage groups as provided herein are preferred. In various embodiments, phosphorothioate linkage groups are preferred, particularly for the gap region (B) of gapmers. In certain embodiments, phosphorothioate linkages are used to link together monomers in the flanking regions (A and C). In various embodiments, phosphorothioate linkages are used for linking regions A or C to region D, and for linking together monomers within region D.

In various embodiments, regions A, B and C, comprise linkage groups other than phosphorothioate, such as phosphodiester linkages, particularly, for instance when the use of nucleoside analogues protects the linkage groups within regions A and C from endo-nuclease degradation—such as when regions A and C comprise LNA monomers.

In various embodiments, adjacent monomers of the oligomer are linked to each other by means of phosphorothioate groups.

It is recognised that the inclusion of phosphodiester linkages, such as one or two linkages, into an oligomer with a phosphorothioate backbone, particularly with phosphorothioate linkage groups between or adjacent to nucleoside

analogue monomers (typically in region A and/or C), can modify the bioavailability and/or bio-distribution of an oligomer—see WO2008/053314, hereby incorporated by reference.

In some embodiments, such as the embodiments referred to above, where suitable and not specifically indicated, all remaining linkage groups are either phosphodiester or phosphorothioate, or a mixture thereof.

In some embodiments all the internucleoside linkage groups are phosphorothioate.

When referring to specific gapmer oligonucleotide sequences, such us those provided herein, it will be understood that, in various embodiments, when the linkages are phosphorothioate linkages, alternative linkages, such as those disclosed herein may be used, for example phosphate (phosphodiester) linkages may be used, particularly for linkages between nucleoside analogues, such as LNA monomers. Likewise, in various embodiments, when referring to specific gapmer oligonucleotide sequences, such as those provided herein, when one or more monomers in region C comprises a 5-methylcytosine base, other monomers in that region may contain unmodified cytosine bases.

Target Nucleic Acid

The terms "nucleic acid" and "polynucleotide" are used interchangeably herein, and are defined as a molecule formed by covalent linkage of two or more monomers, as above-described. Including 2 or more monomers, "nucleic acids" may be of any length, and the term is generic to "oligomers", which have the lengths described herein. The terms "nucleic acid" and "polynucleotide" include single-stranded, double-stranded, partially double-stranded, and circular molecules.

The term "target nucleic acid", as used herein, refers to DNA or RNA (e.g., mRNA or pre-mRNA) encoding a mammalian androgen receptor polypeptide, such as human androgen receptor, such as the nucleic acid having the sequence shown in SEQ ID NO: 1, and naturally occurring allelic variants of such nucleic acids. In certain embodiments, the mammalian androgen receptor is a mouse androgen receptor. In some embodiments, for example when used in research or diagnostics, the "target nucleic acid" is a cDNA or a synthetic oligonucleotide derived from the above DNA or RNA nucleic acid targets. The oligomers according to the invention are typically capable of hybridising to the target nucleic acid.

Exemplary target nucleic acids include mammalian androgen receptor-encoding nucleic acids having the GenBank Accession numbers shown in the table below, along with their corresponding protein sequences:

	GenBank Accession Number Nucleic acid (mRNA/cDNA sequence)	GenBank Accession Number Polypeptide (deduced)
Human	NM_000044	NP_000035
Mouse	NM_013476	NP_038504
Rhesus monkey	NM_001032911	NP_001028083

It is recognised that the above-disclosed GenBank Accession numbers for nucleic acids refer to cDNA sequences and not to mRNA sequences per se. The sequence of a mature mRNA can be derived directly from the corresponding cDNA sequence with thymine bases (T) being replaced by uracil bases (U).

The term "naturally occurring variant thereof" refers to variants of the androgen receptor polypeptide or nucleic acid sequence which exist naturally within the defined taxonomic group, such as mammalian, such as mouse, monkey, and preferably human AR. Typically, when referring to "naturally occurring variants" of a polynucleotide the term also encompasses any allelic variant of the androgen receptor encoding genomic DNA which is found at the Chromosome X: 66.68-66.87 Mb by chromosomal translocation or duplication, and the RNA, such as mRNA derived therefrom. "Naturally occurring variants" may also include variants derived from alternative splicing of the androgen receptor mRNA. When referenced to a specific polypeptide sequence, e.g., the term also includes naturally occurring forms of the protein which may therefore be processed, e.g. by co- or post-translational modifications, such as signal peptide cleavage, proteolytic cleavage, glycosylation, etc.

It is recognised that the human androgen receptor gene exhibits allelic variations that are associated with disease phenotypes (Mooney et al, NAR 15; 31(8) 2003). For example, a $(CAG)_n$ repeat expansion is associated with polyglutamine expansion disorder. Other characterised allelic variants include a $(GGC)_n$ trinucleotide repeat and single nucleotide polymorphisms R726L, T887A and L710H, of which the latter two single nucleotide polymorphisms have been shown to be correlated to enhanced promiscuity of the AR receptor for other steroid ligands. In one embodiment "n" ranges from 5-31. CAG repeats of less than 22 have been associated with an enhanced risk of prostate cancer in African American males.

In various embodiments, the target nucleic acid is an AR allelic variant which comprises a $(CAG)_n$ trinucleotide repeat, or $(GGC)_n$ trinucleotide repeat. In other embodiments, the target nucleic acid is an AR allelic variant which comprises one or more single nucleotide polymorphisms, including R726L, T887A and L710H.

In certain embodiments, oligomers described herein bind to a region of the target nucleic acid (the "target region") by either Watson-Crick base pairing, Hoogsteen hydrogen bonding, or reversed Hoogsteen hydrogen bonding, between the monomers of the oligomer and monomers of the target nucleic acid. Such binding is also referred to as "hybridisation." Unless otherwise indicated, binding is by Watson-Crick pairing of complementary bases (i.e., adenine with thymine (DNA) or uracil (RNA), and guanine with cytosine), and the oligomer binds to the target region because the sequence of the oligomer is identical to, or partially-identical to, the sequence of the reverse complement of the target region; for purposes herein, the oligomer is said to be "complementary" or "partially complementary" to the target region, and the percentage of "complementarity" of the oligomer sequence to that of the target region is the percentage "identity" to the reverse complement of the sequence of the target region.

Unless otherwise made clear by context, the "target region" herein will be the region of the target nucleic acid having the sequence that best aligns with the reverse complement of the sequence of the specified oligomer (or region thereof), using the alignment program and parameters described herein below.

In determining the degree of "complementarity" between oligomers of the invention (or regions thereof) and the target region of the nucleic acid which encodes mammalian androgen receptor, such as those disclosed herein, the degree of "complementarity" (also, "homology") is expressed as the percentage identity between the sequence of the oligomer (or region thereof) and the reverse complement of the sequence of the target region that best aligns therewith. The percentage is calculated by counting the number of aligned bases that are identical as between the 2 sequences, dividing by the total number of contiguous monomers in the oligomer, and multiplying by 100. In such a comparison, if gaps exist, it is

preferable that such gaps are merely mismatches rather than areas where the number of monomers within the gap differs between the oligomer of the invention and the target region.

Amino acid and polynucleotide alignments, percentage sequence identity, and degree of complementarity may be determined for purposes of the invention using the ClustalW algorithm using standard settings: see http://www.ebi.ac.uk/emboss/align/index.html, Method: EMBOSS::water (local): Gap Open=10.0, Gap extend=0.5, using Blosum 62 (protein), or DNAfull for nucleotide/nucleobase sequences.

As will be understood, depending on context, "mismatch" refers to a non-identity in sequence (as, for example, between the nucleobase sequence of an oligomer and the reverse complement of the target region to which it binds; as for example, between the base sequence of two aligned AR encoding nucleic acids), or to noncomplementarity in sequence (as, for example, between an oligomer and the target region to which it binds).

The androgen receptor is known to regulate the expression of several genes, such as a gene selected from the group consisting of Protein kinase C delta (PRKCD), Glutathione S— transferase theta 2 (GSTT2), transient receptor potential cation channel subfamily V member 3 (TRPV3), Pyrroline-5-carboxylate reductase 1 (PYCR1) and ornithine aminotransferase (OAT). Such genes regulated by AR are referred to herein as "androgen receptor (AR) target genes". In various embodiments, the oligomers according to the invention are capable of inhibiting (such as, by down-regulating) the expression of one or more AR target genes in a cell which is expressing, or is capable of expressing (i.e. by alleviating AR repression of the AR target gene in a cell) an AR target gene.

The oligomers which target the androgen receptor mRNA, may hybridize to any site along the target mRNA nucleic acid, such as the 5' untranslated leader, exons, introns and 3' untranslated tail. However, it is preferred that the oligomers which target the androgen receptor mRNA hybridise to the mature mRNA form of the target nucleic acid.

Suitably, the oligomer of the invention or conjugate thereof is capable of down-regulating expression of the androgen receptor gene. In various embodiments, the oligomer (or conjugate) of the invention can effect the inhibition of androgen receptor, typically in a mammalian cell, such as a human cell. In certain embodiments, the oligomers of the invention, or conjugates thereof, bind to the target nucleic acid and effect inhibition of AR mRNA expression of at least 10% or 20% compared to the expression level immediately prior to dosing of the oligomer, more preferably of at least 30%, 40%, 50%, 60%, 70%, 80%, 90% or 95% as compared to the AR expression level immediately prior to dosing of the oligomer. In some embodiments, such inhibition is seen when using from about 0.04 nM to about 25 nM, such as from about 0.8 nM to about 20 nM of the oligomer or conjugate.

In various embodiments, the inhibition of mRNA expression is less than 100% (i.e., less than complete inhibition of expression), such as less than 98% inhibition, less than 95% inhibition, less than 90% inhibition, less than 80% inhibition, such as less than 70% inhibition. In various embodiments, modulation of gene expression can be determined by measuring protein levels, e.g. by the methods such as SDS-PAGE followed by western blotting using suitable antibodies raised against the target protein. Alternatively, modulation of expression levels can be determined by measuring levels of mRNA, e.g. by northern blotting or quantitative RT-PCR. When measuring via mRNA levels, the level of down-regulation when using an appropriate dosage, such as from about 0.04 nM to about 25 nM, such as from about 0.8 nM to about 20 nM, is, in various embodiments, typically to a level of 10-20% of the normal levels in the absence of the compound or conjugate of the invention.

The invention therefore provides a method of down-regulating or inhibiting the expression of the androgen receptor protein and/or mRNA in a cell which is expressing the androgen receptor protein and/or mRNA, the method comprising contacting the cell with an effective amount of the oligomer or conjugate according to the invention to down-regulate or inhibit the expression of the androgen receptor protein and/or mRNA in the cell. Suitably the cell is a mammalian cell, such as a human cell. The contacting may occur, in some embodiments, in vitro. The contacting may occur, in some embodiments, in vivo.

Oligomer Sequences

In some embodiments, the oligomers of the invention have sequences that are identical to a sequence selected from the group consisting of SEQ ID NOS: 2-22. Target regions in human AR mRNA (cDNA) that bind to the oligomers having sequences as set forth in SEQ ID NOs: 2-22 are shown in FIG. **4** (bold and underlined, with the corresponding oligomer SEQ ID NOs indicated above).

Further provided are target nucleic acids (e.g., DNA or mRNA encoding AR) that contain target regions that are complementary or partially-complementary to one or more of the oligomers of the invention. In certain embodiments, the oligomers bind to variants of AR target regions, such as allelic variants (such as an AR gene present at gene locus Xq11-12). In some embodiments, a variant of an AR target region has at least 60%, more preferably at least 70%, more preferably at least 80%, more preferably at least 85%, more preferably at least 90%, more preferably at least 91%, at least 92%, at least 93%, at least 94%, at least 95% sequence identity to the target region in wild-type AR. Thus, in other embodiments, the oligomers of the invention have sequences that differ in 1, 2 or 3 bases when compared to a sequence selected from the group consisting of SEQ ID NOs: 2-22. Typically, an oligomer of the invention that binds to a variant of an AR target region is capable of inhibiting (e.g., by down-regulating) AR.

In other embodiments, oligomers of the invention are LNA oligomers, for example, those oligomers having the sequences shown in SEQ ID NOs: 44-80. In various embodiments, the oligomers of the invention are potent inhibitors of androgen receptor mRNA and protein expression. In various embodiments, oligomers of the invention are LNA oligomers having the sequences of SEQ ID NO: 58 or SEQ ID NO: 77.

In various embodiments, the oligomer comprises or consists of a region having a base sequence which is identical or partially identical to the sequence of the reverse complement of a target region in SEQ ID NO: 1. In various embodiments, the oligomer comprises or consists of a region having a sequence selected from the group consisting of SEQ ID NOS: 2-22 and 86-106.

In certain embodiments, the oligomer comprises or consists of a region having a base sequence which is fully complementary (perfectly complementary) to a target region of a nucleic acid which encodes a mammalian androgen receptor.

However, in some embodiments, the oligomer includes 1, 2, 3, or 4 (or more) mismatches as compared to the best-aligned target region of an AR target nucleic acid, and still sufficiently binds to the target region to effect inhibition of AR mRNA or protein expression. The destabilizing effect of mismatches on Watson-Crick hydrogen-bonded duplex may, for example, be compensated by increased length of the oligomer and/or an increased number of nucleoside analogues, such as LNA monomers, present within the oligomer.

In various embodiments, the oligomer base sequence comprises no more than 3, such as no more than 2 mismatches compared to the base sequence of the best-aligned target region of, for example, a target nucleic acid which encodes a mammalian androgen receptor.

In some embodiments, the oligomer base sequence comprises no more than a single mismatch when compared to the base sequence of the best-aligned target region of a nucleic acid which encodes a mammalian androgen receptor.

In various embodiments, the base sequence of the oligomer of the invention, or of a first region thereof, is preferably at least 80% identical to a base sequence selected from the group consisting of SEQ ID NOS: 2-22 and 86-106, such as at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96% identical, such as 100% identical.

In certain embodiments, the base sequence of the oligomer of the invention or of a first region thereof is at least 80% identical to the base sequence of the reverse complement of a target region present in SEQ ID NO: 1, such as at least 85%, at least 90%, at least 91%, at least 92% at least 93%, at least 94%, at least 95%, at least 96% identical, at least 97% identical, at least 98% identical, at least 99% identical, such as 100% identical.

In various embodiments, the base sequence of the oligomer of the invention, or of a first region thereof, is preferably at least 80% complementary to a target region of SEQ ID NO: 1, such as at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96% complementary, at least 97% complementary, at least 98% complementary, at least 99% complementary, such as 100% complementary (perfectly complementary).

In some embodiments the oligomer (or a first region thereof) has a base sequence selected from the group consisting of SEQ ID NOs: 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, and 22, or is selected from the group consisting of at least 10 contiguous monomers of SEQ ID NOs: 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, and 22. In other embodiments, the sequence of the oligomer of the invention or a first region thereof comprises one, two, or three base moieties that differ from those in oligomers having sequences of SEQ ID NOs: 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, or 22, or the sequences of at least 10 contiguous monomers thereof, when optimally aligned with the selected sequence or region thereof.

In some embodiments the oligomer (or a first region thereof) has a base sequence selected from the group consisting of SEQ ID NOs: 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105 and 106, or the sequences of at least 10 contiguous monomers thereof. In other embodiments, the sequence of the oligomer (or a first region thereof) comprises one, two, or three base moieties that differ from those in oligomers having sequences of SEQ ID NOs: 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105 or 106, or the sequences of at least 10 contiguous monomers thereof, when optimally aligned with the selected sequence or region thereof.

In various embodiments, the oligomers comprise a region of 12, 13, 14, 15 or 16 contiguous monomers having a base sequence identically present in a sequence selected from the group consisting of SEQ ID No 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, and 22. In other embodiments, the oligomers include a region which comprises one, two, or three base moieties that differ from those in oligomers having sequences of SEQ ID NOs: 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, or 22.

In some embodiments the region consists of 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, or 29 contiguous monomers, such as 12-22, such as 12-18 monomers. Suitably, in some embodiments, the region is of the same length as the oligomer of the invention.

In some embodiments the oligomer comprises additional monomers at the 5' or 3' ends, such as, independently, 1, 2, 3, 4 or 5 additional monomers at the 5' end and/or the 3' end of the oligomer, which are non-complementary to the target region. In various embodiments, the oligomer of the invention comprises a region that is complementary to the target, which is flanked 5' and/or 3' by additional monomers. In some embodiments the additional 5' or 3' monomers are nucleosides, such as DNA or RNA monomers. In various embodiments, the 5' or 3' monomers represent region D as referred to in the context of gapmer oligomers herein.

In certain embodiments, the oligomer according to the invention consists of OT comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO:2, such as SEQ ID NO: 44, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID No: 3, such as SEQ ID NO: 45, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 4, such as SEQ ID NO: 46, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 5, such as SEQ ID NO: 47, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 6, such as SEQ ID NOs: 48, 49 or 50, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 7, such as SEQ ID NOs: 51, 52, or 53, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 8, such as SEQ ID NOs: 54, 55 or 56, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 9, such as SEQ ID NO: 57, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 10,

such as SEQ ID NOs: 58, 59, or 60, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 11, such as SEQ ID NO: 61, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 12, such as SEQ ID NO: 62, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 13, such as SEQ ID NOs: 63, 64 or 65, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 14, such as SEQ ID NO: 66, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 15, such as SEQ ID NO: 67, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 16, such as SEQ ID NO: 68, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 17, such as SEQ ID NOs: 69, 70 or 71, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 18, such as SEQ ID NO: 72, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 19, such as SEQ ID NOs: 73, 74 or 75, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 20, such as SEQ ID NO: 76, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 21, such as SEQ ID NOs: 77, 78 or 79, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

In certain embodiments, the oligomer according to the invention consists of or comprises contiguous monomers having a nucleobase sequence according to SEQ ID NO: 22, such as SEQ ID NO: 80, or according to a region of at least 10 contiguous monomers thereof, such as 11, 12, 13, 14, 15 or 16 contiguous monomers thereof.

Nucleosides and Nucleoside Analogues

In various embodiments, at least one of the monomers present in the oligomer is a nucleoside analogue that contains a modified base, such as a base selected from 5-methylcytosine, isocytosine, pseudoisocytosine, 5-bromouracil, 5-propynyluracil, 6-aminopurine, 2-aminopurine, inosine, diaminopurine, 2-chloro-6-aminopurine, xanthine and hypoxanthine.

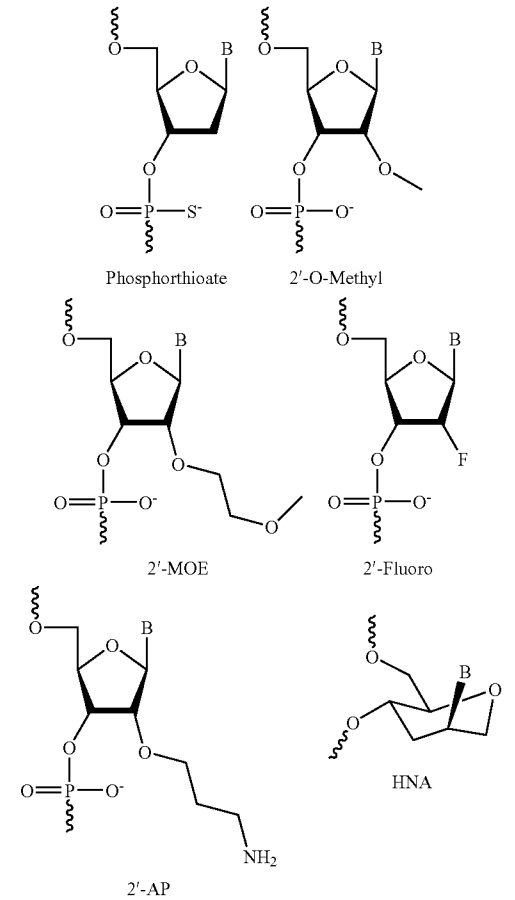
In various embodiments, at least one of the monomers present in the oligomer is a nucleoside analogue that contains a modified sugar.

In some embodiments, the linkage between at least 2 contiguous monomers of the oligomer is other than a phosphodiester linkage.

In certain embodiments, the oligomer includes at least one monomer that has a modified base, at least one monomer (which may be the same monomer) that has a modified sugar, and at least one inter-monomer linkage that is non-naturally occurring.

Specific examples of nucleoside analogues are described by e.g. Freier & Altmann; *Nucl. Acid Res.,* 1997, 25, 4429-4443 and Uhlmann; *Curr. Opinion in Drug Development,* 2000, 3(2), 293-213, and in Scheme 1 (in which some nucleoside analogues are shown as nucleotides):

Scheme 1



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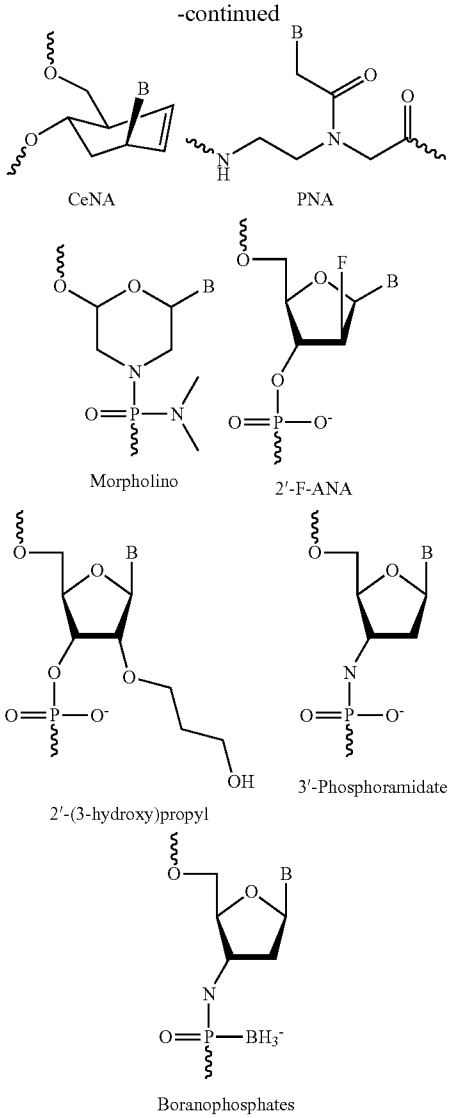


The oligomer may thus comprise or consist of a simple sequence of naturally occurring nucleosides—preferably DNA monomers, but also possibly RNA monomers, or a combination of nucleosides and one or more nucleoside analogues. In some embodiments, such nucleoside analogues suitably enhance the affinity of the oligomer for the target region of the target nucleic acid.

Examples of suitable and preferred nucleoside analogues are described in PCT/DK2006/000512, or are referenced therein.

In some embodiments, the nucleoside analogue comprises a sugar moiety modified to provide a 2'-substituent group, such as 2'-O-alkyl-ribose sugars, 2'-amino-deoxyribose sugars, and 2'-fluoro-deoxyribose sugars.

In some embodiments, the nucleoside analogue comprises a sugar in which a bridged structure, creating a bicyclic sugar (LNA), which enhances binding affinity and may also provide some increased nuclease resistance. In various embodiments, the LNA monomer is selected from oxy-LNA (such as beta-D-oxy-LNA, and alpha-L-oxy-LNA), and/or amino-LNA (such as beta-D-amino-LNA and alpha-L-amino-LNA) and/or thio-LNA (such us beta-D-thio-LNA and alpha-L-thio-LNA) and/or ENA (such as beta-D-ENA and alpha-L-ENA). In certain embodiments, the LNA monomers are beta-D-oxy-LNA. LNA monomers are further described below.

In various embodiments, incorporation of affinity-enhancing nucleoside analogues in the oligomer, such as LNA monomers or monomers containing 2'-substituted sugars, or incorporation of modified linkage groups provides increased nuclease resistance. In various embodiments, incorporation of affinity-enhancing nucleoside analogues allows the size of the oligomer to be reduced, and also reduces the size of the oligomer that binds specifically to a target region of a target sequence.

In some embodiments, the oligomer comprises at least 2 nucleoside analogues. In some embodiments, the oligomer comprises from 3-8 nucleoside analogues, e.g. 6 or 7 nucleoside analogues. In various embodiments, at least one of the nucleoside analogues is a locked nucleic acid (LNA) monomer; for example at least 3 or at least 4, or at least 5, or at least 6, or at least 7, or 8, nucleoside analogues are LNA monomers. In some embodiments, all the nucleoside analogues are LNA monomers.

It will be recognised that when referring to a preferred oligomer base sequence, in certain embodiments, the oligomers comprise a corresponding nucleoside analogue, such as a corresponding LNA monomer or other corresponding nucleoside analogue, which raise the duplex stability (T_m) of the oligomer/target region duplex (i.e. affinity enhancing nucleoside analogues).

In various embodiments, any mismatches (i.e., non-complementarities) between the base sequence of the oligomer and the base sequence of the target region, if present, are preferably located other than in the regions of the oligomer that contain affinity-enhancing nucleoside analogues (e.g., regions A or C), such as within region 13 as referred to herein, and/or within region D as referred to herein, and/or in regions consisting of DNA monomers, and/or in regions which are 5' or 3' to the region of the oligomer that is complementary to the target region.

In some embodiments the nucleoside analogues present within the oligomer of the invention (such as in regions A and C mentioned herein) are independently selected from, for example: monomers containing 2'-O-alkyl-ribose sugars, monomers containing 2'-amino-deoxyribose sugars, monomers containing 2'-fluoro-deoxyribose sugars, LNA monomers, monomers containing arabinose sugars ("ANA monomers"), monomers containing 2'-fluoro-arabinose sugars, monomers containing d-arabino-hexitol sugars ("HNA monomers"), intercalating monomers as defined in Christensen (2002) Nucl. Acids. Res. 30: 4918-4925, hereby incorporated by reference, and 2'-O-methoxyethyl-ribose (2'MOE) sugars. In some embodiments, there is only one of the above types of nucleoside analogues present in the oligomer of the invention, or region thereof.

In certain embodiments, the nucleoside analogues contain 2'MOE sugars, 2% fluoro-deoxyribose sugars, or LNA sugars, and as such the oligonucleotide of the invention may comprise nucleoside analogues which are independently selected from these three types. In certain oligomer embodiments containing nucleoside analogues, at least one of said nucleoside analogues contains a 2'-MOE-ribose sugar, such as 2, 3, 4, 5, 6, 7, 8, 9 or 10 nucleoside analogues containing 2'-MOE-ribose sugars. In some embodiments, at least one nucleoside analogue contains a 2'-fluoro-deoxyribose sugar, such as 2, 3, 4, 5, 6, 7, 8, 9 or 10 nucleoside analogues containing 2'-fluoro-DNA nucleotide sugars.

In various embodiments, the oligomer according to the invention comprises at least one Locked Nucleic Acid (LNA) monomer, such as 1, 2, 3, 4, 5, 6, 7, or 8 LNA monomers, such

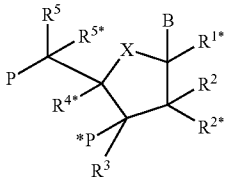
as 3-7 or 4 to 8 LNA monomers, or 3, 4, 5, 6 or 7 LNA monomers. In various embodiments, all the nucleoside analogues are LNA monomers. In certain embodiments, the oligomer comprises both beta-D-oxy-LNA monomers, and one or more of the following LNA monomers: thio-LNA monomers, amino-LNA monomers, oxy-LNA monomers, and/or ENA monomers in either the beta-D or alpha-L configurations, or combinations thereof. In certain embodiments, the cytosine base moieties of all LNA monomers in the oligomer are 5-methylcytosines. In certain embodiments of the invention, the oligomer comprises both LNA and DNA monomers. Typically, the combined total of LNA and DNA monomers is 10-25, preferably 10-20, even more preferably 12-16. In some embodiments of the invention, the oligomer or region thereof consists of at least one LNA monomer, and the remaining monomers are DNA monomers. In certain embodiments, the oligomer comprises only LNA monomers and nucleosides (such as RNA or DNA monomers, most preferably DNA monomers) optionally with modified linkage groups such as phosphorothioate.

In various embodiments, at least one of the nucleoside analogues present in the oligomer has a modified base selected from the group consisting of 5-methylcytosine, isocytosine, pseudoisocytosine, 5-bromouracil, 5-propynyluracil, 6-aminopurine, 2-aminopurine, inosine, diaminopurine, and 2-chloro-6-aminopurine.

LNA

The term "LNA monomer" refers to a nucleoside analogue containing a bicyclic sugar (an "LNA sugar"). The terms "LNA oligonucleotide" and "LNA oligomer" refer to an oligomer containing one or more LNA monomers.

The LNA used in the oligonucleotide compounds of the invention preferably has the structure of the general formula I:



wherein X is selected from —O—, —S—, —N(R^N*), —C(R^6R^6*)—;

B is selected from hydrogen, optionally substituted C_{1-4}-alkoxy, optionally substituted C_{1-4}-alkyl, optionally substituted C_{1-4}-acyloxy, nucleobases, DNA intercalators, photochemically active groups, thermochemically active groups, chelating groups, reporter groups, and ligands;

P designates the radical position for an internucleoside linkage to a succeeding monomer, or a 5'-terminal group, such internucleoside linkage or 5'-terminal group optionally including the substituent R^5 or equally applicable the substituent R^5*;

P* designates an internucleoside linkage to a preceding monomer, or a 3'-terminal group;

R^4* and R^2* together designate a biradical consisting of 1-4 groups/atoms selected from —C(R^aR^b)—, —C(R^a)=C(R^b)—, —C(R^a)=N—, —O—, —Si(R^a)$_2$—, —N(R)—, and >C=Z,

wherein Z is selected from —O—, —S—, and —N(R^a)—, and R^a and R^b each is independently selected from hydrogen, optionally substituted C_{1-12}-alkyl, optionally substituted C_{2-12}-alkenyl, optionally substituted C_{1-12}-alkynyl, hydroxy, C_{2-12}-alkoxyalkyl, C_{2-12}-alkenyloxy, carboxy, C_{1-12}-alkoxycarbonyl, C_{1-12}-alkylcarbonyl, formyl, aryl, aryloxy-carbonyl, aryloxy, arylcarbonyl, heteroaryl, heteroaryloxy-carbonyl, heteroaryloxy, heteroarylcarbonyl, amino, mono- and di(C_{1-6}-alkyl)amino, carbamoyl, mono- and di(C_{1-4}-alkyl)-amino-carbonyl, amino-C_{1-6}-alkyl-aminocarbonyl, mono- and di(C_{1-6}-alkyl)amino-C_{1-6}-alkyl-aminocarbonyl, C_{1-6}-alkyl-carbonylamino, carbamido, C_{1-6}-alkanoyloxy, sulphono, C_{1-6}-alkylsulphonyloxy, nitro, azido, sulphanyl, halogen, DNA intercalators, photochemically active groups, thermochemically active groups, chelating groups, reporter groups, and ligands, where aryl and heteroaryl may be optionally substituted and where two geminal substituents R^a and R^b together may designate optionally substituted methylene (=CH_2), and

each of the substituents R^1*, R^2, R^3, R^5, R^5*, R^6 and R^6*, which are present is independently selected from hydrogen, optionally substituted C_{1-12}-alkyl, optionally substituted C_{2-12}-alkenyl, optionally substituted C_{2-12}-alkynyl, hydroxy, C_{1-12}-alkoxy, C_{2-12}-alkoxyalkyl, C_{2-12}-alkenyloxy, carboxy, C_{1-12}-alkoxycarbonyl, C_{1-12}-alkylcarbonyl, formyl, aryl, aryloxy-carbonyl, aryloxy, arylcarbonyl, heteroaryl, heteroaryloxy-carbonyl, heteroaryloxy, heteroarylcarbonyl, amino, mono- and di(C_{1-6}-alkyl)amino, carbamoyl, mono- and di(C_{1-6}-alkyl)-amino-carbonyl, amino-C_{1-6}-alkyl-aminocarbonyl, mono- and di(C_{1-6}-alkyl)amino-C_{1-6}-alkyl-aminocarbonyl, C_{1-6}-alkyl-carbonylamino, carbamido, alkanoyloxy, sulphono, C_{1-6}-alkylsulphonyloxy, nitro, azido, sulphanyl, C_{1-6}-alkylthio, halogen, DNA intercalators, photochemically active groups, thermochemically active groups, chelating groups, reporter groups, and ligands, where aryl and heteroaryl may be optionally substituted, and where two geminal substituents together may designate oxo, thioxo, imino, or optionally substituted methylene, or together may form a spiro biradical consisting of a 1-5 carbon atom(s) alkylene chain which is optionally interrupted and/or terminated by one or more heteroatoms/groups selected from —O—, —S—, and —(NR^N)— where R^N is selected from hydrogen and C_{1-4}-alkyl, and where two adjacent (non-geminal) substituents may designate an additional bond resulting in a double bond; and R^N*, when present and not involved in a biradical, is selected from hydrogen and C_{1-4}-alkyl; and basic salts and acid addition salts thereof;

In some embodiments, R^5* is selected from H, —CH_3, —CH_2—CH_3, —CH_2—O—CH_3, and —CH=CH_2.

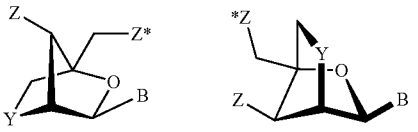
In various embodiments, R^4* and R^2* together designate a biradical selected from —C(R^aR^b)—O—, —C(R^aR^b)—C(R^cR^d)—O—, —C(R^aR^b)—C(R^cR^d)—C(R^eR^f)—O—, —C(R^aR^b)—O—C(R^cR^d)—, —C(R^aR^b)—O—C(R^cR^d)—O—, —C(R^aR^b)—C(R^cR^d)—, —C(R^aR^b)—C(R^cR^d)—C(R^eR^f)—, —C(R^a)=C(R^b)—C(R^cR^d)—, —C(R^aR^b)—N(R^c—, —C(R^aR^b)—C(R^cR^d)—N(R^c)—, —C(R^aR^b)—N(R^c)—O—, and —C(R^aR^b)—S—, —C(R^aR^b)—C(R^cR^d)—S—, wherein R^a, R^b, R^c, R^d, R^e, and R^f each is independently selected from hydrogen, optionally substituted C_{1-12}-alkyl, optionally substituted C_{2-12}-alkenyl, optionally substituted C_{2-12}-alkynyl, hydroxy, C_{1-12}-alkoxyalkyl, C_{2-12}-alkenyloxy, carboxy, C_{1-12}-alkoxycarbonyl, C_{1-12}-alkylcarbonyl, formyl, aryl, aryloxy-carbonyl, aryloxy, arylcarbonyl, heteroaryl, heteroaryloxy-carbonyl, heteroaryloxy, heteroarylcarbonyl, amino, mono- and di(C_{1-6}-alkyl)amino, carbamoyl, mono- and di(C_{1-6}-alkyl)-amino-carbonyl, amino-C_{1-6}-alkyl-aminocarbonyl, mono- and di(C_{1-6}-alkyl)amino-C_{1-6}-alkyl-aminocarbonyl, C_{1-6}-alkyl-carbonylamino, carbamido, C_{1-6}-alkanoyloxy, sulphono, C_{1-6}-alkylsulphonyloxy, nitro, azido, sulphanyl, C_{1-6}-alkylthio, halogen, DNA intercalators, photochemically active groups, thermochemically active

groups, chelating groups, reporter groups, and ligands, where aryl and heteroaryl may be optionally substituted and where two geminal substituents R^a and R^b together may designate optionally substituted methylene (=CH_2),

In a further embodiment R^{4*} and R^{2*} together designate a biradical (bivalent group) selected from —CH_2—O—, —CH_2—S—, —CH_2—NH—, —CH_2—N(CH_3)—, —CH_2—CH_2—O—, —CH_2—CH(CH_3)—, —CH_2—CH_2—S—, —CH_2—CH_2—NH—, —CH_2—CH_2—CH_2—, —CH_2—CH_2—CH_2—O—, —CH_2—CH_2—CH(CH_3)—, —CH=CH—CH_2—, —CH_2—O—CH_2—O—, —CH_2—NH—O—, —CH_2—N(CH_3)—O—, —CH_2—O—CH_2—, —CH(CH_3)—O—, —CH(CH_2—O—CH_3)—O—.

For all chiral centers, asymmetric groups may be found in either R or S orientation.

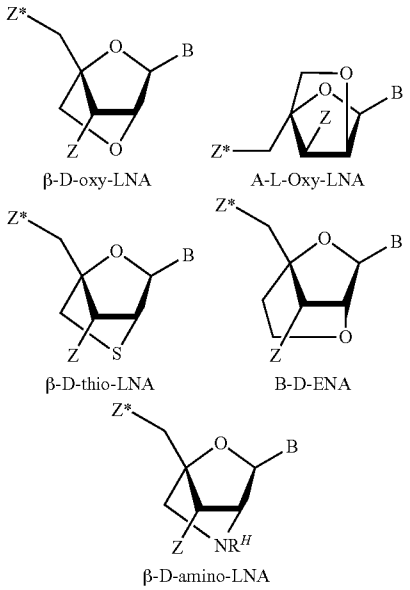
Preferably, the LNA monomer used in the oligomer of the invention comprises at least one LNA monomer according to any of the formulas



wherein Y is —O—, —O—CH2-, —S—, —NH—, or N(RH); Z and Z* are independently selected among an internucleotide linkage, a terminal group or a protecting group; B constitutes a natural or non-natural nucleotide base moiety, and R^H is selected from hydrogen and C_{1-4}-alkyl.

Specifically preferred LNA monomers are shown in Scheme 2:

Scheme 2



The term "thio-LNA" refers to an LNA monomer in which Y in the general formula above is selected from S or —CH_2—S—. Thio-LNA can be in either the beta-D or alpha-L-configuration.

The term "amino-LNA" refers to an LNA monomer in which Y in the general formula above is selected from —N(H)—, N(R)—, CH_2—N(H)—, and —CH_2—N(R)— where R is selected from hydrogen and C_{1-4}-alkyl. Amino-LNA can be hi either the beta-D or alpha-L-configuration.

The term "oxy-LNA" refers to an LNA monomer in which Y in the general formula above represents —O— or —CH_2—O—. Oxy-LNA can be in either the beta-D or alpha-L-configuration.

The term "ENA" refers to an LNA monomer in which Y in the general formula above is —CH_2—O— (where the oxygen atom of —CH_2—O— is attached to the 2'-position relative to the base B).

In various embodiments, the LNA monomer is selected from a beta-D-oxy-LNA monomer, and alpha-L-oxy-LNA monomer, a beta-D-amino-LNA monomer, and beta-D-thio-LNA monomer, in particular a beta-D-oxy-LNA monomer.

In the present context, the term "C_{1-4}alkyl" means a linear or branched saturated hydrocarbon chain wherein the chain has from one to four carbon atoms, such as methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl, sec-butyl and tert-butyl.

RNAse H Recruitment

In some embodiments, an oligomer functions via non-RNase-mediated degradation of a target mRNA, such as by steric hindrance of translation, or other mechanisms; however, in various embodiments, oligomers of the invention are capable of recruiting an endo-ribonuclease (RNase), such as RNase H.

Typically, the oligomer, comprises a region of at least 6, such as at least 7 contiguous monomers, such as at least 8 or at least 9 contiguous monomers, including 7, 8, 9, 10, 11, 12, 13, 14, 15 or 16 contiguous monomers, which, when forming a duplex with the target region of the target RNA, is capable of recruiting RNase. The region of the oligomer which is capable of recruiting RNAse may be region B, as referred to in the context of a gapmer as described herein. In some embodiments, the region of the oligomer which is capable of recruiting RNAse, such as region B, consists of 10, 11, 12, 13, 14, 15, 16, 17, 18, 19 or 20 monomers.

EP 1 222 309 provides in vitro methods for determining RNaseH activity, which may be used to determine the ability of the oligomers of the invention to recruit RNaseH. An oligomer is deemed capable of recruiting RNaseH if, when contacted with the complementary region of the RNA target, it has an initial rate, as measured in pmol/l/min, of at least 1%, such as at least 5%, such as at least 10% or less than 20% of an oligonucleotide having the same base sequence but containing only DNA monomers, with no 2' substitutions, with phosphorothioate linkage groups between all monomers in the oligonucleotide, using the methodology provided by Examples 91-95 of EP 1 222 309, incorporated herein by reference.

In some embodiments, an oligomer is deemed essentially incapable of recruiting RNaseH if, when contacted with the complementary target region of the RNA target, and RNaseH, the RNaseH initial rate, as measured in pmol/l/min, is less than 1%, such as less than 5%, such as less than 10% or less than 20% of the initial rate determined using an oligonucleotide having the same base sequence, but containing only DNA monomers, with no 2' substitutions, with phosphorothioate linkage groups between all monomers in the oligonucleotide, using the methodology provided by Examples 91-95 of EP 1 222 309.

In other embodiments, an oligomer is deemed capable of recruiting RNaseH if, when contacted with the complementary target region of the RNA target, and RNaseH, the RNaseH initial rate, as measured in pmol/l/min, is at least 20%, such as at least 40%, such as at least 60%, such as at

least 80% of the initial rate determined using an oligonucleotide having the same base sequence, but containing only DNA monomers, with no 2' substitutions, with phosphorothioate linkage groups between all monomers in the oligonucleotide, using the methodology provided by Examples 91-95 of EP 1 222 309.

Typically, the region of the oligomer which forms the duplex with the complementary target region of the target RNA and is capable of recruiting RNase contains DNA monomers and LNA monomers and forms a DNA/RNA-like duplex with the target region. The LNA monomers are preferably in the alpha-L configuration, particularly preferred being alpha-L-oxy LNA.

In various embodiments, the oligomer of the invention comprises both nucleosides and nucleoside analogues, and is in the form of a gapmer, a headmer or a mixmer.

A "headmer" is defined as an oligomer that comprises a first region and a second region that is contiguous thereto, with the 5'-most monomer of the second region linked to the 3'-most monomer of the first region. The first region comprises a contiguous stretch of non-RNase recruiting nucleoside analogues and the second region comprises a contiguous stretch (such as at least 7 contiguous monomers) of DNA monomers or nucleoside analogue monomers recognizable and cleavable by the RNase

A "tailmer" is defined as an oligomer that comprises a first region and a second region that is contiguous thereto, with the 5'-most monomer of the second region linked to the 3'-most monomer of the first region. The first region comprises a contiguous stretch (such as at least 7 contiguous monomers) of DNA monomers or nucleoside analogue monomers recognizable and cleavable by the RNase, and the second region comprises a contiguous stretch of non-RNase recruiting nucleoside analogues.

Other "chimeric" oligomers, called "mixmers", consist of an alternating composition of (i) DNA monomers or nucleoside analogue monomers recognizable and cleavable by RNase, and (ii) non-RNase recruiting nucleoside analogue monomers.

In some embodiments, in addition to enhancing affinity of the oligomer for the target region, some nucleoside analogues also mediate RNase (e.g., RNaseH) binding and cleavage. Since •-L-LNA monomers recruit RNaseH activity to a certain extent, in some embodiments, gap regions (e.g., region B as referred to herein) of oligomers containing •-L-LNA monomers consist of fewer monomers recognizable and cleavable by the RNaseH, and more flexibility in the mixmer construction is introduced.

Conjugates

In the context of this disclosure, the term "conjugate" indicates a compound formed by the covalent attachment ("conjugation") of an oligomer as described herein, to one or more moieties that are not themselves nucleic acids or monomers ("conjugated moieties"). Examples of such conjugated moieties include macromolecular compounds such as proteins, fatty acid chains, sugar residues, glycoproteins, polymers, or combinations thereof. Typically proteins may be antibodies for a target protein. Typical polymers may be polyethylene glycol.

Accordingly, provided herein are conjugates comprising an oligomer as herein described, and at least one conjugated moiety that is not a nucleic acid or monomer, covalently attached to said oligomer. Therefore, in certain embodiments where the oligomer of the invention consists of contiguous monomers having a specified sequence of bases, as herein disclosed, the conjugate may also comprise at least one conjugated moiety that is covalently attached to the oligomer.

In various embodiments of the invention, the oligomer is conjugated to a moiety that increases the cellular uptake of oligomeric compounds. WO2007/031091 provides suitable ligands and conjugates, which are hereby incorporated by reference.

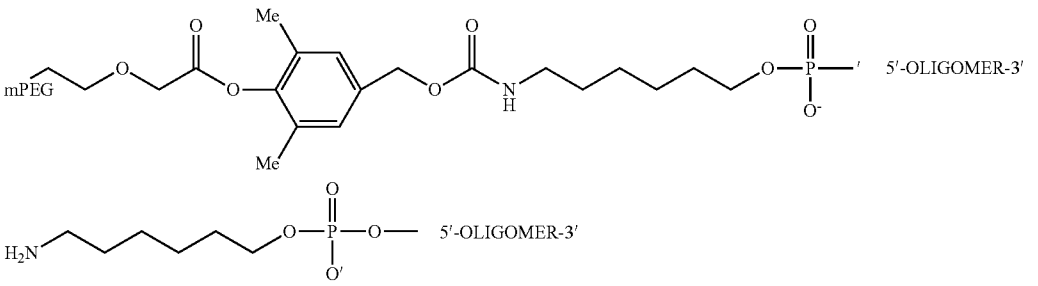
In various embodiments, conjugation (to a conjugated moiety) may enhance the activity, cellular distribution or cellular uptake of the oligomer of the invention. Such moieties include, but are not limited to, antibodies, polypeptides, lipid moieties such as a cholesterol moiety, cholic acid, a thioether, e.g. Hexyl-s-tritylthiol, a thiocholesterol, an aliphatic chain, e.g., dodecandiol or undecyl residues, a phospholipids, e.g., di-hexadecyl-rac-glycerol or triethylammonium 1,2-di-o-hexadecyl-rac-glycero-3-h-phosphonate, a polyamine or a polyethylene glycol chain, an adamantane acetic acid, a palmityl moiety, an octadecylamine or hexylamino-carbonyl-oxycholesterol moiety.

In certain embodiments, the oligomers of the invention are conjugated to active drug substances, for example, aspirin, ibuprofen, a sulfa drug, an antidiabetic, an antibacterial or an antibiotic.

In certain embodiments the conjugated moiety is a sterol, such as cholesterol.

In various embodiments, the conjugated moiety comprises or consists of a positively charged polymer, such as a positively charged peptides of, for example 1-50, such as 2-20 such as 3-10 amino acid residues in length, and/or polyalkylene oxide such as polyethylene glycol (PEG) or polypropylene glycol—see WO 2008/034123, hereby incorporated by reference. Suitably the positively charged polymer, such as a polyalkylene oxide may be attached to the oligomer of the invention via a linker such as the releasable linker described in WO 2008/034123.

By way of example, the following moieties may be used in the conjugates of the invention:



Activated Oligomers

The term "activated oligomer," as used herein, refers to an oligomer of the invention that is covalently linked (i.e., functionalized) to at least one functional moiety that permits covalent linkage of the oligomer to one or more conjugated moieties, i.e., moieties that are not themselves nucleic acids or monomers, to form the conjugates herein described. Typically, a functional moiety will comprise a chemical group that is capable of covalently bonding to the oligomer via, e.g., a 3'-hydroxyl group or the exocyclic NH_2 group of the adenine base, a spacer that is preferably hydrophilic and a terminal group that is capable of binding to a conjugated moiety (e.g., an amino, sulfhydryl or hydroxyl group). In some embodiments, this terminal group is not protected, e.g., is an NH, group. In other embodiments, the terminal group is protected, for example, by any suitable protecting group such as those described in "Protective Groups in Organic Synthesis" by Theodora W. Greene and Peter G. M. Wuts, 3rd edition (John Wiley & Sons, 1999). Examples of suitable hydroxyl protecting groups include esters such as acetate ester, aralkyl groups such as benzyl, diphenylmethyl, or triphenylmethyl, and tetrahydropyranyl. Examples of suitable amino protecting groups include benzyl, alpha-methylbenzyl, diphenylmethyl, triphenylmethyl, benzyloxycarbonyl, tert-butoxycarbonyl, and acyl groups such as trichloroacetyl or trifluoroacetyl.

In some embodiments, the functional moiety is self-cleaving. In other embodiments, the functional moiety is biodegradable. See e.g., U.S. Pat. No. 7,087,229, which is incorporated by reference herein in its entirety.

In some embodiments, oligomers of the invention are functionalized at the 5' end in order to allow covalent attachment of the conjugated moiety to the 5' end of the oligomer. In other embodiments, oligomers of the invention can be functionalized at the 3' end. In still other embodiments, oligomers of the invention can be functionalized along the backbone or on the heterocyclic base moiety. In yet other embodiments, oligomers of the invention can be functionalized at more than one position independently selected from the 5' end, the 3' end, the backbone and the base.

In some embodiments, activated oligomers of the invention are synthesized by incorporating during the synthesis one or more monomers that is covalently attached to a functional moiety. In other embodiments, activated oligomers of the invention are synthesized with monomers that have not been functionalized, and the oligomer is functionalized upon completion of synthesis.

In some embodiments, the oligomers are functionalized with a hindered ester containing an aminoalkyl linker, wherein the alkyl portion has the formula $(CH_2)_w$, wherein w is an integer ranging from 1 to 10, preferably about 6, wherein the alkyl portion of the alkylamino group can be straight chain or branched chain, and wherein the functional group is attached to the oligomer via an ester group (—O—C(O)—$(CH_2)_w$NH).

In other embodiments, the oligomers are functionalized with a hindered ester containing a $(CH_2)_w$-sulfhydryl (SH) linker, wherein w is an integer ranging from 1 to 10, preferably about 6, wherein the alkyl portion of the alkylamino group can be straight chain or branched chain, and wherein the functional group attached to the oligomer via an ester group (—O—C(O)—$(CH_2)_w$SH) In some embodiments, sulfhydryl-activated oligonucleotides are conjugated with polymer moieties such as polyethylene glycol or peptides (via formation of a disulfide bond).

Activated oligomers containing hindered esters as described above can be synthesized by any method known in the art, and in particular, by methods disclosed in PCT Publication No. WO 2008/034122 and the examples therein, which is incorporated herein by reference in its entirety.

Activated oligomers covalently linked to at least one functional moiety can be synthesized by any method known in the art, and in particular, by methods disclosed in U.S. Patent Publication No. 2004/0235773, which is incorporated herein by reference in its entirety, and in Zhao et al. (2007) J. Controlled Release 119:143-152; and Zhao et al. (2005) Bioconjugate Chem. 16:758-766.

In still other embodiments, the oligomers of the invention are functionalized by introducing sulfhydryl, amino or hydroxyl groups into the oligomer by means of a functionalizing reagent substantially as described in U.S. Pat. Nos. 4,962,029 and 4,914,210, i.e., a substantially linear reagent having a phosphoramidite at one end linked through a hydrophilic spacer chain to the opposing end which comprises a protected or unprotected sulfhydryl, amino or hydroxyl group. Such reagents primarily react with hydroxyl groups of the oligomer. In some embodiments, such activated oligomers have a functionalizing reagent coupled to a 5'-hydroxyl group of the oligomer. In other embodiments, the activated oligomers have a functionalizing reagent coupled to a 3'-hydroxyl group. In still other embodiments, the activated oligomers of the invention have a functionalizing reagent coupled to a hydroxyl group on the backbone of the oligomer. In yet further embodiments, the oligomer of the invention is functionalized with more than one of the functionalizing reagents as described in U.S. Pat. Nos. 4,962,029 and 4,914,210, incorporated herein by reference in their entirety. Methods of synthesizing such functionalizing reagents and incorporating them into monomers or oligomers are disclosed in U.S. Pat. Nos. 4,962,029 and 4,914,210.

In some embodiments, the 5'-terminus of a solid-phase bound oligomer is functionalized with a dienyl phosphoramidite derivative, followed by conjugation of the deprotected oligomer with, e.g., an amino acid or peptide via a Diels-Alder cycloaddition reaction.

In various embodiments, the incorporation of monomers containing 2'-sugar modifications, such as a 2'-carbamate substituted sugar or a 2'-(O-pentyl-N-phthalimido)-deoxyribose sugar into the oligomer facilitates covalent attachment of conjugated moieties to the sugars of the oligomer. In other embodiments, an oligomer with an amino-containing linker at the 2'-position of one or more monomers is prepared using a reagent such as, for example, 5'-dimethoxytrityl-2'-O-(e-phthalimidylaminopentyl)-2'-deoxyadenosine-3'-N,N-diisopropyl-cyanoethoxy phosphoramidite. See, e.g., Manoharan, et al., Tetrahedron Letters, 1991, 34, 7171.

In still further embodiments, the oligomers of the invention have amine-containing functional moieties on the nucleobase, including on the N6 purine amino groups, on the exocyclic N2 of guanine, or on the N4 or 5 positions of cytosine. In various embodiments, such functionalization may be achieved by using a commercial reagent that is already functionalized in the oligomer synthesis.

Some functional moieties are commercially available, for example, heterobifunctional and homobifunctional linking moieties are available from the Pierce Co. (Rockford, Ill.). Other commercially available linking groups are 5'-Amino-Modifier C6 and 3'-Amino-Modifier reagents, both available from Glen Research Corporation (Sterling, Va.). 5'-Amino-Modifier C6 is also available from ABI (Applied Biosystems Inc., Foster City, Calif.) as Aminolink-2, and 3'-Amino-Modifier is also available from Clontech Laboratories Inc. (Palo Alto, Calif.).

Compositions

In various embodiments, the oligomer of the invention is used in pharmaceutical formulations and compositions. Suitably, such compositions comprise a pharmaceutically acceptable diluent, carrier, salt or adjuvant. PCT/DK2006/000512 provides suitable and preferred pharmaceutically acceptable diluents, carriers and adjuvants—which are hereby incorporated by reference. Suitable dosages, formulations, administration routes, compositions, dosage forms, combinations with other therapeutic agents, pro-drug formulations are also provided in PCT/DK2006/000512—which are also hereby incorporated by reference. Details on techniques for formulation and administration also may be found in the latest edition of "REMINGTON'S PHARMACEUTICAL SCIENCES" (Maack Publishing Co, Easton Pa.).

In some embodiments, an oligomer of the invention is covalently linked to a conjugated moiety to aid in delivery of the oligomer across cell membranes. An example of a conjugated moiety that aids in delivery of the oligomer across cell membranes is a lipophilic moiety, such as cholesterol. In various embodiments, an oligomer of the invention is formulated with lipid formulations that form liposomes, such as Lipofectamine 2000 or Lipofectamine RNAiMAX, both of which are commercially available from Invitrogen. In some embodiments, the oligomers of the invention are formulated with a mixture of one or more lipid-like non-naturally occurring small molecules ("lipidoids"). Libraries of lipidoids can be synthesized by conventional synthetic chemistry methods and various amounts and combinations of lipidoids can be assayed in order to develop a vehicle for effective delivery of an oligomer of a particular size to the targeted tissue by the chosen route of administration. Suitable lipidoid libraries and compositions can be found, for example in Akinc et al. (2008) Nature Biotechnol., available at http://www.nature.corn/nbt/journal/vaop/ncurrent/abs/nbt1402.html, which is incorporated by reference herein.

As used herein, the term "pharmaceutically acceptable salts" refers to salts that retain the desired biological activity of the herein identified compounds and exhibit acceptable levels of undesired toxic effects. Non-limiting examples of such salts can be formed with organic amino acid and base addition salts formed with metal cations such as zinc, calcium, bismuth, barium, magnesium, aluminum, copper, cobalt, nickel, cadmium, sodium, potassium, and the like, or with a cation formed from ammonia, N,N'-dibenzylethylenediamine, D-glucosamine, tetraethylammonium, or ethylenediamine; or (c) combinations of (a) and (b); e.g., a zinc tannate salt or the like.

In certain embodiments, the pharmaceutical compositions according to the invention comprise other active ingredients in addition to an oligomer or conjugate of the invention, including active agents useful for the treatment of cancer, such as prostate cancer or breast cancer, particularly agents used in conventional antiandrogen therapy.

In some embodiments, additional active agents are non-steroidal antiandrogens (NSAAs), which block the binding of androgens at the receptor site, or luteinizing hormone-releasing hormone analogues (LHRH-As) that suppress testicular production of androgens to castrate levels.

NSAAs such as CASODEX, when used with an LHRH-A as part of Combined Androgen Blockade therapy, help to inhibit the growth of prostate cancer cells. In one embodiment, the invention provides for a combined androgen blockade therapy, characterised in that the therapy comprises administering the pharmaceutical composition according to the invention, and an NSAA and/or LHRH-A agent, which in certain embodiments are administered prior to, during or subsequent to the administration of the pharmaceutical compositions of the invention.

The invention also provides a kit of parts wherein a first part comprises at least one oligomer, conjugate and/or the pharmaceutical composition according to the invention and a further part comprises a non-steroidal antiandrogen and/or a luteinizing hormone-releasing hormone analogue. It is therefore envisaged that the kit of parts may be used in a method of treatment, as referred to herein, where the method comprises administering both the first part and the further part, either simultaneously or one after the other.

Applications

The term "treatment" as used herein refers to both treatment of an existing disease (e.g., a disease or disorder as referred to herein below), or prevention of a disease, i.e., prophylaxis. It will therefore be recognised that, in certain embodiments, "treatment" includes prophylaxis.

In various embodiments, the oligomers of the invention may be utilized as research reagents for, for example, diagnostics, therapeutics and prophylaxis.

In some embodiments, such oligomers may be used for research purposes to specifically inhibit the expression of androgen receptor protein (typically by degrading or inhibiting the AR mRNA and thereby preventing protein formation) in cells and experimental animals, thereby facilitating functional analysis of the target or an appraisal of its usefulness as a target for therapeutic intervention.

In certain embodiments, the oligomers may be used in diagnostics to detect and quantitate androgen receptor expression in cells and tissues by Northern blotting, in-situ hybridisation or similar techniques.

In various therapeutic embodiments, a non-human animal or a human suspected of having a disease or disorder which can be treated by modulating the expression of androgen receptor is treated by administering an effective amount of an oligomer in accordance with this invention. Further provided are methods of treating a mammal, such as treating a human, suspected of having or being prone to a disease or condition, associated with expression of androgen receptor by administering a therapeutically or prophylactically effective amount of one or more of the oligomers, conjugates or compositions of the invention.

In certain embodiments, the invention also provides for the use of the compounds or conjugates of the invention as described for the manufacture of a medicament for the treatment of a disorder as referred to herein, or for a method of the treatment of a disorder as referred to herein.

In various embodiments, the invention also provides for a method for treating a disorder as referred to herein, said method comprising administering a compound according to the invention as herein described, and/or a conjugate according to the invention, and/or a pharmaceutical composition according to the invention to a patient in need thereof.

Medical Indications

In certain therapeutic embodiments, the disorder to be treated is cancer, such as prostate cancer or breast cancer. In various embodiments, the treatment of such a disease or condition according to the invention may be combined with one or more other anti-cancer treatments, such as radiotherapy, chemotherapy or immunotherapy.

In certain other embodiments, the disorder to be treated is selected from alopecia, benign prostatic hyperplasia, spinal and muscular atrophy and Kennedy disease and polyglutamate disease.

In various embodiments, the disease or disorder is associated with a mutation of the AR gene or a gene whose protein

product is associated with or interacts with AR. Therefore, in various embodiments, the target snRNA is a mutated form of the AR sequence, for example, it comprises one or more single point mutations or triplet repeats.

In other embodiments, the disease or disorder is associated with abnormal levels of a mutated form of androgen receptor. In various embodiments, the disease or disorder is associated with abnormal levels of a wild-type form of AR.

In various embodiments, the invention relates to methods of modulating the expression of the gene product of an androgen receptor target gene, i.e., a gene that is regulated by AR. Such AR receptor target gene products are selected form the group consisting of Protein kinase C delta (PRKCD), Glutathione S-transferase theta 2 (GSTT2), transient receptor potential cation channel subfamily V member 3 (TRPV3), Pyrroline-5-carboxylate reductase 1 (PYCR1) and ornithine aminotransferase (OAT). In some embodiments, modulation of an AR target gene results in increased expression or activity of the target gene. In other embodiments, modulation of an AR target gene results in decreased expression or activity of the target gene.

The invention further provides use of a compound of the invention in the manufacture of a medicament for the treatment of any and all conditions disclosed herein.

In various embodiments, the invention is directed to a method of treating a mammal suffering from or susceptible to a condition associated with abnormal levels of androgen receptor mRNA or protein, comprising administering to the mammal a therapeutically effective amount of an oligomer of the invention, or a conjugate thereof, that comprises one or more LNA monomers.

An interesting aspect of the invention is directed to the use of an oligomer (compound) as defined herein or a conjugate as defined herein for the preparation of a medicament for the treatment of a condition as disclosed herein above.

In various embodiments, the invention encompasses a method of preventing or treating a disease comprising administering a therapeutically effective amount of an oligomer according to the invention, or a conjugate thereof, to a human in need of such therapy.

In certain embodiments, the LNA oligomers of the invention, or conjugates thereof, are administered for a short period time rather than continuously.

In certain embodiments of the invention, the oligomer (compound) is linked to a conjugated moiety, for example, in order to increase the cellular uptake of the oligomer. In one embodiment the conjugated moiety is a sterol, such as cholesterol.

In various embodiments, the invention is directed to a method for treating abnormal levels of androgen receptor, the method comprising administering an oligomer of the invention, or a conjugate or a pharmaceutical composition thereof, to a patient in need of such treatment, and further comprising the administration of a further chemotherapeutic agent. In some embodiments, the chemotherapeutic agent is conjugated to the oligomer, is present in the pharmaceutical composition, or is administered in a separate formulation.

The invention also relates to an oligomer, a composition or a conjugate as defined herein for use as a medicament.

The invention further relates to use of a compound, composition, or a conjugate as defined herein for the manufacture of a medicament for the treatment of abnormal levels of androgen receptor or expression of mutant forms of AR (such as allelic variants, such as those associated with one of the diseases referred to herein).

Moreover, in various embodiments, the invention relates to a method of treating a subject suffering from a disease or condition selected from cancer, such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease, the method comprising the step of administering a pharmaceutical composition as defined herein to the subject in need thereof.

Suitable dosages, formulations, administration routes, compositions, dosage forms, combinations with other therapeutic agents, pro-drug formulations are also provided in PCT/DK2006/000512—which is hereby incorporated by reference.

The invention also provides for a pharmaceutical composition comprising a compound or a conjugate as herein described or a conjugate, and a pharmaceutically acceptable diluent, carrier or adjuvant. PCT/DK2006/000512 provides suitable and preferred pharmaceutically acceptable diluents, carriers and adjuvants—which are hereby incorporated by reference.

EMBODIMENTS

The following embodiments of the invention may be used in combination with the other embodiments described herein.

1. An oligomer of between 10-50 nucleobases in length which comprises a contiguous nucleobase sequence of a total of between 10-50 nucleobases, wherein said contiguous nucleobase sequence is at least 80% homologous to a corresponding region of a nucleic acid which encodes a mammalian androgen receptor.

2. The oligomer according to embodiment 1, wherein said oligomer comprises at least one LNA unit.

3. The oligomer according to embodiment 1 or 2, wherein the contiguous nucleobase sequence comprises no more than 3, such as no more than 2 mismatches to the corresponding region of a nucleic acid which encodes a mammalian androgen receptor.

4. The oligomer according to embodiment 3, wherein said contiguous nucleobase sequence comprises no more than a single mismatch to the corresponding region of a nucleic acid which encodes a mammalian androgen receptor.

5. The oligomer according to embodiment 4, wherein said contiguous nucleobase sequence comprises no mismatches, (i.e. is complementary to) the corresponding region of a nucleic acid which encodes a mammalian androgen receptor.

6. The oligomer according to any one of embodiments 1-5, wherein the nucleobase sequence of the oligomer consists of the contiguous nucleobase sequence.

7. The oligomer according to any one of embodiments 1-6, wherein the nucleic acid which encodes a mammalian androgen receptor is the human androgen receptor nucleotide sequence such as SEQ ID No 1, or a naturally occurring allelic variant thereof.

8. The oligomer according to any one of embodiments 1-7, wherein the contiguous nucleobase sequence is complementary to a corresponding region of both the human androgen receptor nucleic acid sequence and a non-human mammalian androgen receptor nucleic acid sequence, such as the mouse androgen receptor nucleic acid sequence.

9. The oligomer according to any one of embodiments 1 to 8, wherein the contiguous nucleobase sequence comprises a contiguous subsequence of at least 7, nucleobase residues which, when formed in a duplex with the complementary androgen receptor target RNA is capable of recruiting RNaseH.

10. The oligomer according to embodiment 9, wherein the contiguous nucleobase sequence comprises of a contiguous subsequence of at least 8, at least 9 or at least 10 nucleobase

residues which, when formed in a duplex with the complementary androgen receptor target RNA is capable of recruiting RNaseH.

11. The oligomer according to any one of embodiments 9 or 10 wherein said contiguous subsequence is at least 9 or at least 10 nucleobases in length, such as at least 12 nucleobases or at least 14 nucleobases in length, such as 14, 15 or 16 nucleobases residues which, when formed in a duplex with the complementary androgen receptor target RNA is capable of recruiting RNaseH.

12. The oligomer according to embodiment any one of embodiments 1-11 wherein said oligomer is conjugated with one or more non-nucleobase compounds.

13. The oligomer according to any one of embodiments 1-12, wherein said oligomer has a length of between 10-22 nucleobases.

14. The oligomer according to any one of embodiments 1-13, wherein said oligomer has a length of between 12-18 nucleobases.

15. The oligomer according to any One of embodiments 1-14, wherein said oligomer has a length of 14, 15 or 16 nucleobases.

16. The oligomer according to any one of embodiments 1-15, wherein said continuous nucleobase sequence corresponds to a contiguous nucleotide sequence present in a nucleic acid sequence selected from the group consisting of SEQ ID NO 86-106.

17. The oligomer according to any one of embodiments 1-16, wherein the oligomer or contiguous nucleobase sequence comprises, or is selected from a corresponding nucleobase sequence present in a nucleotide sequence selected from the group consisting of SEQ ID NO 2-22.

18. The oligomer according to any one of embodiments 1-17, wherein said contiguous nucleobase sequence comprises at least one affinity enhancing nucleotide analogue.

19. The oligomer according to embodiment 18, wherein said contiguous nucleobase sequence comprises a total of 2, 3, 4, 5, 6, 7, 8, 9 or 10 affinity enhancing nucleotide analogues, such as between 5 and 8 affinity enhancing nucleotide analogues.

20. The oligomer according to any one of embodiments 1-19 which comprises at least one affinity enhancing nucleotide analogue, wherein the remaining nucleobases are selected from the group consisting of DNA nucleotides and RNA nucleotides, preferably DNA nucleotides.

21. The oligomer according to any one of embodiments 1-20, wherein the oligomer comprises of a sequence of nucleobases of formula, in 5' to 3' direction, A-B-C, and optionally of formula A-B-C-D, wherein:

(a) consists or comprises of at least one nucleotide analogue, such as 1, 2, 3, 4, 5 or 6 nucleotide analogues, preferably between 2-5 nucleotide analogues, preferably 2, 3 or 4 nucleotide analogues, most preferably 2, 3 or 4 consecutive nucleotide analogues and;

(b) consists or comprises at least five consecutive nucleobases which are capable of recruiting RNAseH (when formed in a duplex with a complementary RNA molecule, such as the AR mRNA target), such as DNA nucleobases, such as 5, 6, 7, 8, 9, 10, 11 or 12 consecutive nucleobases which are capable of recruiting RNAseH, or between 6-10, or between 7-9, such as 8 consecutive nucleobases which are capable of recruiting RNAseH, and;

(c) consists or comprises of at least one nucleotide analogue, such as 1, 2, 3, 4, 5, or 6 nucleotide analogues, preferably between 2-5 nucleotide analogues, such as 2, 3 or 4 nucleotide analogues, most preferably 2, 3 or 4 consecutive nucleotide analogues, and;

(d) when present, consists or comprises, preferably consists, of one or more DNA nucleotide, such as between 1-3 or 1-2 DNA nucleotides.

22. The oligomer according to embodiment 21, wherein region A consists or comprises of 2, 3 or 4 consecutive nucleotide analogues.

23. The oligomer according to any one of embodiments 21-22, wherein region B consists or comprises of 7, 8, 9 or 10 consecutive DNA nucleotides or equivalent nucleobases which are capable of recruiting RNAseH when formed in a duplex with a complementary RNA, such as the androgen receptor mRNA target.

24. The oligomer according to any one of embodiments 21-23, wherein region C consists or comprises of 2, 3 or 4 consecutive nucleotide analogues.

25. The oligomer according to any one of embodiments 21-24, wherein region D consists, where present, of one or two DNA nucleotides.

26. The oligomer according to any one of embodiments 21-25, wherein:

(a) Consists or comprises of 3 contiguous nucleotide analogues;

(b) Consists or comprises of 7, 8, 9 or 10 contiguous DNA nucleotides or equivalent nucleobases which are capable of recruiting RNAseH when formed in a duplex with a complementary RNA, such as the androgen receptor mRNA target;

(c) Consists or comprises of 3 contiguous nucleotide analogues;

(d) Consists, where present, of one or two DNA nucleotides.

27. The oligomer according to embodiment 26, wherein the contiguous nucleobase sequence consists of 10, 11, 12, 13 or 14 nucleobases, and wherein;

(a) Consists of 1, 2 or 3 contiguous nucleotide analogues;

(b) Consists of 7, 8, or 9 consecutive DNA nucleotides or equivalent nucleobases which are capable of recruiting RNAseH when formed in a duplex with a complementary RNA, such as the androgen receptor mRNA target;

(c) Consists of 1, 2 or 3 contiguous nucleotide analogues;

(d) Consists, where present, of one DNA nucleotide.

28. The oligomer according to anyone of embodiments 21-27, wherein B comprises at least one LNA nucleobase which is in the alpha-L configuration, such as alpha-L-oxy LNA.

29. The oligomer according to any one of embodiments 1-28, wherein the nucleotide analogue(s) are independently or collectively selected from the group consisting of: Locked Nucleic Acid (LNA) units; 2'-O-alkyl-RNA units, 2'-OMe-RNA units, 2'-amino-DNA units, 2'-fluoro-DNA units, PNA units, HNA units, and INA units.

30. The oligomer according to embodiment 29 wherein all the nucleotide analogues(s) are LNA units.

31. The oligomer according to any one of embodiments 1-30, which comprises 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10 LNA units such as between 2 and 8 nucleotide LNA units.

32. The oligomer according to any one of the embodiments 29-31, wherein the LNAs are independently selected from oxy-LNA, thio-LNA, and amino-LNA, in either of the beta-D and alpha-L configurations or combinations thereof.

33. The oligomer according to embodiment 32, wherein the LNAs are all beta-D-oxy-LNA.

34. The oligomer according to any one of embodiments 21-33, wherein the nucleotide analogues or nucleobases of regions A and C are beta-D-oxy-LNA.

35. The oligomer according to any one of embodiments 1-34, wherein at least one of the nucleobases present in the oligomer is a modified nucleobase selected from the group consisting of 5-methylcytosine, isocytosine, pseudoisocytosine, 5-bromouracil, 5-propynyluracil, 6-aminopurine, 2-aminopurine, inosine, diaminopurine, and 2-chloro-6-aminopurine.

36. The oligomer according to any one of embodiments 1-35, wherein said oligomer hybridises with a corresponding mammalian androgen receptor mRNA with a T_m of at least 50° C.

37. The oligomer according to any one of embodiments 1-36, wherein said oligomer hybridises with a corresponding mammalian androgen receptor mRNA with a T_m, of no greater than 80° C.

38. The oligomer according to any one of embodiments 1-37, wherein the internucleoside linkages are independently selected from the group consisting of: phosphodiester, phosphorothioate and boranophosphate.

39. The oligomer according to embodiment 38, wherein the oligomer comprises at least one phosphorothioate internucleoside linkage.

40. The oligomer according to embodiment 39, wherein the internucleoside linkages adjacent to or between DNA or RNA units, or within region B are phosphorothioate linkages.

41. The oligomer according to embodiment 39 or 40, wherein the linkages between at least one pair of consecutive nucleotide analogues is a phosphodiester linkage.

42. The oligomer according to embodiment 39 or 40, wherein all the linkages between consecutive nucleotide analogues are phosphodiester linkages.

43. The oligomer according to embodiment 42 wherein all the internucleoside linkages are phosphorothioate linkages.

44. A conjugate comprising the oligomer according to any one of the embodiments 1-43 and at least one non-nucleotide or non-polynucleotide moiety covalently attached to said compound.

45. A pharmaceutical composition comprising an oligomer as defined in any of embodiments 1-43 or a conjugate as defined in embodiment 44, and a pharmaceutically acceptable diluent, carrier, salt or adjuvant.

46. A pharmaceutical composition according to 45, wherein the oligomer is constituted as a pro-drug.

47. A pharmaceutical composition according to embodiment 45 or 46, which further comprises a further therapeutic agent selected from the group consisting of Non-steroidal Antiandrogens and Luteinizing hormone-releasing hormone analogues.

48. Use of an oligomer as defined in any one of the embodiments 1-43, or a conjugate as defined in embodiment 44, for the manufacture of a medicament for the treatment of a disease or disorder selected from the group consisting of: Cancer such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease.

49. An oligomer as defined in any one of the embodiments 1-43, or a conjugate as defined in embodiment 44, for use in the treatment of a disease or disorder selected from the group consisting of: Cancer such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease.

50. A method for treating a disease or disorder selected from the group consisting of: Cancer such as breast cancer or prostate cancer, alopecia, benign prostatic hyperplasia, spinal and muscular atrophy, Kennedy disease and polyglutamate disease, said method comprising administering an oligomer as defined in one of the embodiments 1-43, or a conjugate as defined in embodiment 44, or a pharmaceutical composition as defined in any one of the embodiments 45-47, to a patient in need thereof.

51. A method for treating an cancer such as prostate cancer or breast cancer, said method comprising administering an oligomer as defined in one of the embodiments 1-43, or a conjugate as defined in embodiment 44, or a pharmaceutical composition as defined in any one of the embodiments 45-47, to a patient in need thereof.

52. A method of reducing or inhibiting the expression of androgen receptor in a cell or a tissue, the method comprising the step of contacting said cell or tissue with a compound as defined in one of the embodiments 1-43, or a conjugate as defined in embodiment 44, or a pharmaceutical composition as defined in any one of the embodiments 45-47, so that expression of androgen receptor is reduce or inhibited.

A method for modulating the expression of a gene which is regulated by the androgen receptor (i.e. an androgen receptor target) in a cell which is expressing said gene, said method comprising the step of contacting said cell or tissue with a compound as defined in one of the embodiments 1-43, or a conjugate as defined in embodiment 44, or a pharmaceutical composition as defined in any one of the embodiments 45-47, so that expression of androgen receptor target is modulated.

EXAMPLES

Example 1

Monomer Synthesis

The LNA monomer building blocks and derivatives were prepared following published procedures and references cited therein—see WO07/031,081 and the references cited therein.

Example 2

Oligonucleotide Synthesis

Oligonucleotides were synthesized according to the method described in WO07/031,081. Table 1 shows examples of sequences of antisense oligonucleotides of the invention. Tables 2 and 3 show examples of antisense oligonucleotides (oligomers) of the invention.

Example 3

Design of the Oligonucleotides

In accordance with the invention, a series of oligomers were designed to target different regions of human androgen receptor mRNA (GenBank Accession number NM_000044; SEQ ID NO: 1).

SEQ ID NOS: 2-22, shown in Table 1, below, are sequences of oligomers designed to target human androgen receptor mRNA. The target region of the target nucleic acid is indicated, in the table.

TABLE 1

Antisense Oligonucleotide Sequences

SEQ ID NO	Sequence (5'-3')	Length (bases)	Target site NM_000044
SEQ ID NO: 2	GAGAACCATCCTCACC	16	1389-1404
SEQ ID NO: 3	GGACCAGGTAGCCTGT	16	1428-1443
SEQ ID NO: 4	CCCCTGGACTCAGATG	16	1881-1896
SEQ ID NO: 5	GCACAAGGAGTGGGAC	16	1954-1969
SEQ ID NO: 6	GCTGTGAAGAGAGTGT	16	2422-2437
SEQ ID NO: 7	TTTGACACAAGTGGGA	16	2663-2678
SEQ ID NO: 8	GTGACACCCAGAAGCT	16	2813-2828
SEQ ID NO: 9	CATCCCTGCTTCATAA	16	2975-2990
SEQ ID NO: 10	ACCAAGTTTCTTCAGC	16	3008-3023
SEQ ID NO: 11	CTTGGCCCACTTGACC	16	3263-3278
SEQ ID NO: 12	TCCTGGAGTTGACATT	16	3384-3399
SEQ ID NO: 13	CACTGGCTGTACATCC	16	3454-3469
SEQ ID NO: 14	CATCCAAACTCTTGAG	16	3490-3505
SEQ ID NO: 15	GCTTTCATGCACAGGA	16	3529-3544
SEQ ID NO: 16	GAAGTTCATCAAAGAA	16	3594-3609
SEQ ID NO: 17	AGTTCCTTGATGTAGT	16	3616-3631
SEQ ID NO: 18	TTGCACAGAGATGATC	16	3809-3824
SEQ ID NO: 19	GATGGGCTTGACTTTC	16	3845-3860
SEQ ID NO: 20	CAGGCAGAAGACATCT	16	3924-3939
SEQ ID NO: 21	CCCAAGGCACTGCAGA	16	3960-3975
SEQ ID NO: 22	GCTGACATTCATAGCC	16	3114-3129
SEQ ID NO: 86	TGGGGAGAACCATCCTCACCCTGC	24	1385-1408
SEQ ID NO: 87	TCCAGGACCAGGTAGCCTGTGGGG	24	1424-1447
SEQ ID NO: 88	TGTTCCCCTGGACTCAGATGCTCC	24	1877-1990
SEQ ID NO: 89	TGGGGCACAAGGAGTGGGACGCAC	24	1950-1973
SEQ ID NO: 90	TTCGGCTGTGAAGAGAGTGTGCCA	24	2418-2441
SEQ ID NO: 91	CGCTTTTGACACAAGTGGGACTGG	24	2659-2682
SEQ ID NO: 92	CATAGTGACACCCAGAAGCTTCAT	24	2809-2832
SEQ ID NO: 93	GAGTCATCCCTGCTTCATAACATT	24	2971-2994
SEQ ID NO: 94	GATTACCAAGTTTCTTCAGCTTCC	24	3004-3027
SEQ ID NO: 95	AGGCCTTGGCCCACTTGACCACGT	24	3259-3282
SEQ ID NO: 96	AGCATCCTGGAGTTGACATTGGTG	24	3380-3403
SEQ ID NO: 97	GACACACTGGCTGTACATCCGGGA	24	3450-3473
SEQ ID NO: 98	GAGCCATCCAAACTCTTGAGAGAG	24	3486-3509
SEQ ID NO: 99	CAGTGCTTTCATGCACAGGAATTC	24	35254548
SEQ ID NO: 100	ATTCGAAGTTCATCAAAGAATTTT	24	3590-3613
SEQ ID NO: 101	ATCGAGTTCCTTGATGTAGTTCAT	24	3612-3635

TABLE 1-continued

Antisense Oligonucleotide Sequences

SEQ ID NO	Sequence (5'-3')	Length (bases)	Target site NM_000044
SEQ ID NO: 102	GCACTTGCACAGAGATGATCTCTG	24	3805-3828
SEQ ID NO: 103	AATAGATGGGCTTGACTTTCCCAG	24	3841-3864
SEQ ID NO: 104	ATAACAGGCAGAAGACATCTGAAA	24	3920-3943
SEQ ID NO: 105	ATTCCCCAAGGCACTGCAGAGGAG	24	3956-3979
SEQ ID NO: 106	ATGGGCTGACATTCATAGCCTTCA	24	3110-3133

In SEQ ID NOs: 23-43, shown below in Table 2, upper case, boldface letters indicate nucleoside analogue monomers (e.g., •-D-oxy LNA monomers) and subscript "s" represents phosphorothiote linkage groups between the monomers. The absence of a subscript "s" (if any) indicates a phosphodiester linkage group. Lower case letters represent DNA monomers.

TABLE 2

Oligonucleotide designs

SEQ ID NO	Sequence (5'-3')
SEQ ID NO: 23	5'-$G_sA_sG_sa_sa_sc_sc_sa_st_sc_sc_st_sc_sA_sC_sC$-3'
SEQ ID NO: 24	5'-$G_sG_sA_sc_sc_sa_sg_sg_st_sa_sg_sc_sc_sT_sG_sT$-3'
SEQ ID NO: 25	5'-$C_sC_sC_sc_st_sg_sg_sa_sc_st_sc_sa_sg_sA_sT_sG$-3'
SEQ ID NO: 26	5'-$G_sC_sA_sc_sa_sa_sg_sg_sa_sg_st_sg_sg_sG_sA_sC$-3'
SEQ ID NO: 27	5'-$G_sC_sT_sg_st_sg_sa_sa_sg_sa_sg_sa_sg_sT_sG_sT$-3'
SEQ ID NO: 28	5'-$T_sT_sT_sG_sa_sc_sa_sc_sa_sa_sg_st_sg_sG_sG_sA$-3'
SEQ ID NO: 29	5'-$G_sT_sG_sa_sc_sa_sc_sc_sc_sa_sg_sa_sa_sG_sC_sT$-3'
SEQ ID NO: 30	5'-$C_sA_sT_sc_sc_sc_st_sg_sc_st_st_sc_sa_sT_sA_sA$-3'
SEQ ID NO: 31	5'-$A_sC_sC_sa_sa_sg_st_st_st_sc_st_st_sc_sA_sG_sC$-3'
SEQ ID NO: 32	5'-$C_sT_sT_sG_sg_sc_sc_sc_sa_sc_st_st_sg_sA_sC_sC$-3'
SEQ ID NO: 33	5'-$T_sC_sC_st_sg_sg_sa_sg_st_st_sg_sa_sc_sA_sT_sT$-3'
SEQ ID NO: 34	5'-$C_sA_sC_st_sg_sg_sc_st_sg_st_sa_sc_sa_sT_sC_sC$-3'
SEQ ID NO: 35	5'-$C_sA_sT_sc_sc_sa_sa_sa_sc_st_sc_st_st_sG_sA_sG$-3'
SEQ ID NO: 36	5'-$G_sC_sT_st_st_sc_sa_st_sg_sc_sa_sc_sa_sG_sG_sA$-3'
SEQ ID NO: 37	5'-$G_sA_sA_sg_st_st_sc_sa_st_sc_sa_sa_sa_sG_sA_sA$-3'
SEQ ID NO: 38	5'-$A_sG_sT_st_sc_sc_st_st_sg_sa_st_sg_st_sA_sG_sT$-3'

TABLE 2-continued

Oligonucleotide designs

SEQ ID NO	Sequence (5'-3')
SEQ ID NO: 39	5'-$T_sT_sG_sc_sa_sc_sa_sg_sg_sa_sg_sg_sa_st_sg_sA_sT_sC$-3'
SEQ ID NO: 40	5'-$G_sA_sT_sg_sg_sg_sc_st_st_sg_sa_sc_st_sT_sT_sC$-3'
SEQ ID NO: 41	5'-$C_sA_sG_sg_sc_sa_sg_sa_sa_sg_sa_sc_sa_sT_sC_sT$-3'
SEQ ID NO: 42	5'-$C_sC_sC_sa_sa_sg_sg_sc_sa_sc_st_sg_sc_sA_sG_sA$-3'
SEQ ID NO: 43	5'-$G_sC_sT_sg_sa_sc_sa_st_st_sc_sa_st_sa_sG_sC_sC$-3'

Example 4

In Vitro Model: Cell Culture

The effect of antisense oligonucleotides on target nucleic acid expression can be tested in any of a variety of cell types provided that the target nucleic acid is present at measurable levels. The target can be expressed endogenously or by transient or stable transfection of a nucleic acid encoding said target. The expression level of target nucleic acid can be routinely determined using, for example, Northern blot analysis, Real-Time PCR, Ribonuclease protection assays. The following cell types are provided for illustrative purposes, but other cell types can be routinely used, provided that the target is expressed in the cell type chosen.

Cells were cultured in the appropriate medium as described below and maintained at 37° C. at 95-98% humidity and 5% CO_2. Cells were routinely passaged 2-3 times weekly.

A549 The human lung cancer cell line A5439 was cultured in DMEM (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 μg/ml).

MCF7 The human breast cancer cell line MCF7 was cultured in EagleMEM (Sigma)+10% fetal bovine serum (PBS)+2 mM Glutamax I+1×NEAA+gentamicin (25 μg/ml).

Example 5

In Vitro Model: Treatment with Antisense Oligonucleotide

The cell lines listed in Example 4 were treated with an oligomer using the cationic liposome formulation LipofectAMINE 2000 (Gibco) as transfection vehicle. Cells were

seeded in 6-well cell culture plates (NUNC) and treated when 80-90% confluent. Oligomer concentrations used ranged from 1 nM to 16 nM final concentration. Formulation of oligomer-lipid complexes were carried out essentially as described by the manufacturer using serum-free OptiMEM (Gibco) and a final lipid concentration of 5 μg/mL LipofectAMINE 2000. Cells were incubated at 37° C. for 4 hours and treatment was stopped by removal of oligomer-containing culture medium. Cells were washed and serum-containing media was added. After oligomer treatment, cells were allowed to recover for 20 hours before they were harvested for RNA analysis.

Example 6

In Vitro Model: Extraction of RNA and cDNA Synthesis

Total RNA Isolation and First Strand Synthesis

Total RNA was extracted from cells transfected as described above and using the Qiagen RNeasy kit (Qiagen cat. no. 74104) according to the manufacturer's instructions. First strand synthesis was performed using Reverse Transcriptase reagents from Ambion according to the manufacturer's instructions.

For each sample, the volume of 0.5 •g total RNA was adjusted to 10.8 •l with RNase free H_2O and mixed with 2 •l random decamers (50 •M) and 4 •l dNTP mix (2.5 mM each dNTP) and heated to 70° C. for 3 min, after which the samples were rapidly cooled on ice. After cooling the samples on ice, 2 •l 10× Buffer RT, 1 •l MMLV Reverse Transcriptase (100 U/•l) and 0.25 •l RNase inhibitor (10 U/•l) were added to each sample, followed by incubation at 42° C. for 60 min, heat inactivation of the enzyme at 95° C. for 10 min and then cooling of the sample to 4° C.

Example 7

In Vitro Model: Analysis of Oligonucleotide Inhibition of Androgen Receptor Expression by Real-Time PCR

Antisense modulation of androgen receptor expression can be assayed in a variety of ways known in the art. For example, androgen receptor mRNA levels can be quantitated by, e.g., Northern blot analysis, competitive polymerase chain reaction (PCR), or real-time PCR. Real-time quantitative PCR is presently preferred. RNA analysis can be performed on total cellular RNA or mRNA.

Methods of RNA isolation and RNA analysis such as Northern blot analysis are routine in the art and are taught in, for example, Current Protocols in Molecular Biology, John Wiley and Sons.

Real-time quantitative (PCR) can be conveniently accomplished using the commercially available Multi-Color Real Time PCR Detection System, available from Applied Biosystems.

Real-Time Quantitative PCR Analysis of Androgen Receptor mRNA Levels

The amount of human androgen receptor mRNA in the samples was quantified using the human androgen receptor ABI Prism Pre-Developed TaqMan Assay Reagents (Applied Biosystems cat. no. Hs00171172_m1) according to the manufacturer's instructions.

Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) mRNA quantity was used as an endogenous control for normalizing any variance in sample preparation.

The amount of human GAPDH mRNA in the samples was quantified using the human GAPDH ABI Prism Pre-Developed TaqMan Assay Reagent (Applied Biosystems cat. no. 4310884E) according to the manufacturer's instructions.

Real-time Quantitative PCR is a technique well known in the art and is taught in for example Heid et al. Real time quantitative PCR, Genome Research (1996), 6: 986-994.

Real Time PCR

The cDNA from the first strand synthesis performed as described in Example 6 was diluted 2-20 times, and analyzed by real time quantitative PCR using Taqman 7500 FAST or 7900 FAST from Applied Biosystems. The primers and probe were mixed with 2× Taqman Fast Universal PCR master mix (2×) (Applied Biosystems Cat. #4364103) and added to 4 μl cDNA to a final volume of 10 μl. Each sample was analysed in duplicate. Standard curves were generated by assaying 2-fold dilutions of a cDNA that had been prepared on material purified from a cell line expressing the RNA of interest. Sterile H_2O was used instead of cDNA for the no-template control. PCR program: 95° C. for 30 seconds, followed by 40 cycles of 95° C., 3 seconds, 60° C., 20-30 seconds. Relative quantities of target mRNA were determined from the calculated Threshold cycle using the Applied Biosystems Fast System SDS Software Version 1.3.1.21. or SDS Software Version 2.3.

Example 8

In Vitro Analysis: Antisense Inhibition of Human Androgen Receptor mRNA Expression by Oligonucleotide Compounds

Oligonucleotides presented in Table 3 were evaluated for their potential to knock down androgen receptor mRNA expression at concentrations of 1, 4 and 16 nM (see FIGS. **1** and **2**).

The data in Table 3 are presented as percentage down-regulation relative to mock transfected cells at 16 nM. Lower case letters represent DNA monomers, bold, upper case letters represent β-D-oxy-LNA monomers. All cytosine bases in the LNA monomers are 5-methylcytosines. Subscript "s" represents a phosphorothioate linkage.

TABLE 3

Inhibition of human androgen receptor mRNA expression by oligonucleotides

Test substance	Sequence (5'-3')	Percent inhibition of Androgen recpetor MCF7	Percent inhibition of Androgen receptor A549
SEQ ID NO: 44	5′-**G$_s$A$_s$G$_s$**a$_s$a$_s$c$_s$c$_s$a$_s$t$_s$c$_s$c$_s$t$_s$c$_s$**A$_s$C$_s$C**-3′	80.1	63.8
SEQ ID NO: 45	5′-**G$_s$G$_s$A$_s$**c$_s$c$_s$a$_s$g$_s$g$_s$t$_s$a$_s$g$_s$c$_s$c$_s$**T$_s$G$_s$T**-3′	89.0	88.2

TABLE 3-continued

Inhibition of human androgen receptor mRNA expression by oligonucleotides

Test substance	Sequence (5'-3')	Percent inhibition of Androgen recpetor MCF7	Percent inhibition of Androgen receptor A549
SEQ ID NO: 46	5'-C$_s$C$_s$C$_s$c$_s$t$_s$g$_s$g$_s$a$_s$c$_s$t$_s$c$_s$a$_s$g$_s$A$_s$T$_s$G-3'	89.4	82.8
SEQ ID NO: 47	5'-G$_s$C$_s$A$_s$c$_s$a$_s$a$_s$g$_s$g$_s$a$_s$g$_s$t$_s$g$_s$g$_s$G$_s$A$_s$C-3'	83.1	77.7
SEQ ID NO: 48	5'-G$_s$C$_s$T$_s$g$_s$t$_s$g$_s$a$_s$a$_s$g$_s$a$_s$g$_s$a$_s$g$_s$T$_s$G$_s$T-3'	93.8	96.7
SEQ ID NO: 49	5'-C$_s$T$_s$G$_s$t$_s$g$_s$a$_s$a$_s$g$_s$a$_s$g$_s$a$_s$g$_s$T$_s$G-3'	n.d.	n.d.
SEQ ID NO: 50	5'-T$_s$G$_s$t$_s$g$_s$a$_s$a$_s$g$_s$a$_s$g$_s$a$_s$G$_s$T$_s$-3'	n.d.	n.d.
SEQ ID NO: 51	5'-T$_s$T$_s$T$_s$g$_s$a$_s$c$_s$a$_s$c$_s$a$_s$a$_s$g$_s$t$_s$g$_s$G$_s$G$_s$A-3'	96.9	95.5
SEQ ID NO: 52	5'-T$_s$T$_s$G$_s$a$_s$c$_s$a$_s$c$_s$a$_s$a$_s$g$_s$t$_s$g$_s$G$_s$G-3'	n.d.	n.d.
SEQ ID NO: 53	5'-T$_s$G$_s$a$_s$c$_s$a$_s$c$_s$a$_s$a$_s$g$_s$t$_s$G$_s$G-3'	n.d.	n.d.
SEQ ID NO: 54	5'-G$_s$T$_s$G$_s$a$_s$c$_s$a$_s$c$_s$c$_s$c$_s$a$_s$g$_s$a$_s$G$_s$C$_s$T-3'	95.4	98.3
SEQ ID NO: 55	5'-T$_s$G$_s$A$_s$c$_s$a$_s$c$_s$c$_s$c$_s$a$_s$g$_s$a$_s$a$_s$G$_s$C-3'	n.d.	n.d.
SEQ ID NO: 56	5'-G$_s$A$_s$c$_s$a$_s$c$_s$c$_s$c$_s$a$_s$g$_s$a$_s$A$_s$G-3'	n.d.	n.d.
SEQ ID NO: 57	5'-C$_s$A$_s$T$_s$c$_s$c$_s$t$_s$g$_s$c$_s$t$_s$t$_s$c$_s$a$_s$T$_s$A$_s$A-3'	89.5	88.9
SEQ ID NO: 58	5'-A$_s$C$_s$C$_s$a$_s$a$_s$g$_s$t$_s$t$_s$t$_s$c$_s$t$_s$t$_s$c$_s$A$_s$G$_s$C-3'	95.6	98.9
SEQ ID NO: 59	5'-C$_s$C$_s$A$_s$a$_s$g$_s$t$_s$t$_s$t$_s$c$_s$t$_s$t$_s$c$_s$A$_s$G-3'	n.d.	n.d.
SEQ ID NO: 60	5'-C$_s$A$_s$a$_s$g$_s$t$_s$t$_s$t$_s$c$_s$t$_s$t$_s$C$_s$A-3'	n.d.	n.d.
SEQ ID NO: 61	5'-C$_s$T$_s$T$_s$g$_s$g$_s$c$_s$c$_s$c$_s$a$_s$c$_s$t$_s$t$_s$g$_s$A$_s$C$_s$C-3'	86.7	93.3
SEQ ID NO: 62	5'-T$_s$C$_s$C$_s$t$_s$g$_s$g$_s$a$_s$g$_s$t$_s$t$_s$g$_s$a$_s$c$_s$A$_s$T$_s$T-3'	81.3	93.0
SEQ ID NO: 63	5'-C$_s$A$_s$C$_s$t$_s$g$_s$g$_s$c$_s$t$_s$g$_s$t$_s$a$_s$c$_s$a$_s$T$_s$C$_s$C-3'	90.9	98.4
SEQ ID NO: 64	5'-A$_s$C$_s$T$_s$g$_s$g$_s$c$_s$t$_s$g$_s$t$_s$a$_s$c$_s$a$_s$T$_s$C-3'	n.d.	n.d.
SEQ ID NO: 65	5'-C$_s$T$_s$g$_s$g$_s$c$_s$t$_s$g$_s$t$_s$a$_s$c$_s$A$_s$T-3'	n.d.	n.d.
SEQ ID NO: 66	5'-C$_s$A$_s$T$_s$c$_s$c$_s$a$_s$a$_s$a$_s$c$_s$t$_s$c$_s$t$_s$t$_s$G$_s$A$_s$G-3'	79.8	95.3
SEQ ID NO: 67	5'-G$_s$C$_s$T$_s$t$_s$t$_s$c$_s$a$_s$t$_s$g$_s$c$_s$a$_s$c$_s$a$_s$G$_s$G$_s$A-3'	83.5	97.0
SEQ ID NO: 68	5'-G$_s$A$_s$A$_s$g$_s$t$_s$t$_s$c$_s$a$_s$t$_s$c$_s$a$_s$a$_s$a$_s$G$_s$A$_s$A-3'	88.2	85.6
SEQ ID NO: 69	5'-A$_s$G$_s$T$_s$t$_s$c$_s$c$_s$t$_s$t$_s$g$_s$a$_s$t$_s$g$_s$t$_s$A$_s$G$_s$T-3'	92.7	94.0

TABLE 3-continued

Inhibition of human androgen receptor mRNA expression by oligonucleotides

Test substance	Sequence (5'-3')	Percent inhibition of Androgen recpetor MCF7	Percent inhibition of Androgen receptor A549
SEQ ID NO: 70	5'-$G_sT_sT_sc_sc_st_st_sg_sa_st_sg_sT_sA_sG$-3'	n.d.	n.d.
SEQ ID NO: 71	5'-$T_sT_sc_sc_st_st_sg_sa_st_sg_sT_sA$-3'	n.d.	n.d.
SEQ ID NO: 72	5'-$T_sT_sG_sc_sa_sc_sa_sg_sa_sg_sa_st_sg_sA_sT_sC$-3'	79.2	90.4
SEQ ID NO: 73	5'-$G_sA_sT_sg_sg_sg_sc_st_st_sg_sa_sc_st_sT_sT_sC$-3'	91.1	97.3
SEQ ID NO: 74	5'-$A_sT_sg_sg_sg_sc_st_st_sg_sa_sc_st_sT_sT$-3'	n.d.	n.d.
SEQ ID NO: 75	5'-$T_sG_sg_sg_sc_st_st_sg_sa_sc_sT_sT$-3'	n.d.	n.d.
SEQ ID NO: 76	5'-$C_sA_sG_sg_sc_sa_sg_sa_sa_sg_sa_sc_sa_sT_sC_sT$-3'	85.9	94.3
SEQ ID NO: 77	5'-$C_sC_sC_sa_sa_sg_sg_sc_sa_sc_st_sg_sc_sA_sG_sA$-3'	93.0	98.5
SEQ ID NO: 78	5'-$C_sC_sA_sa_sg_sg_sc_sa_sc_st_sg_sc_sA_sG$-3'	n.d.	n.d.
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SEQ ID NO: 80	5'-$G_sC_sT_sg_sa_sc_sa_st_st_sc_sa_st_sa_sG_sC_sC$-3'	n.d.	n.d.

As shown in Table 3, oligonucleotides having the sequences set forth in SEQ ID NOs: 48, 51, 54, 58, 63, 69, 73 and 77 at 16 nM demonstrated greater than 90% inhibition of androgen receptor mRNA expression in A549 and MCF7 cells in these experiments.

In certain embodiments, oligomers based on the tested antisense oligomer sequences and designs, but having, for example, different lengths (shorter or longer) and/or monomer content (e.g. the type and/or number of nucleoside analogues) than those shown, e.g., in Table 3, could also provide suitable inhibition of androgen receptor expression.

Example 9

In Vivo Analysis: Antisense Inhibition of Mouse Androgen Receptor mRNA Liver Expression by Oligonucleotide Compounds

Nude mice were dosed i.v. q3dx4 with 100 mg/kg oligonucleotide (group size of 5 mice). The antisense oligonucleotides (SEQ ID:48, SEQ ID:51, SEQ ID:58, SEQ ID:63, SEQ ID:77) were dissolved in phosphate buffered saline. Animals were sacrificed 24 h after last dosing and liver tissue was sampled and stored in RNA later until RNA extraction and QPCR analysis. Total RNA was extracted and AR mRNA expression in liver samples was measured by QPCR as described, in Example 7 using a mouse AR QPCR assay (cat. Mm01238475_m1, Applied Biosystems). Results were normalised to mouse GAPDH (cat. no. 4352339E, Applied Biosystems) and knock-down was quantitated relative to saline treated controls. The data in Table 4 are presented as percentage down-regulation relative to saline treated animals.

TABLE 4

In vivo knock-down of AR mRNA expression

Compound	Liver (% KD)
Saline	0
SEQ ID: 51 100 mg/kg	65.0 +/− 12.6
SEQ ID: 58 100 mg/kg	95.2 +/− 1.0
SEQ ID: 77 100 mg/kg	91.9 +/− 3.9

As shown in Table 4, oligonucleotides of SEQ ID NOs: 58 and 77 at 100 mg/kg demonstrated greater than 90% inhibition of androgen receptor mRNA expression in mouse liver cells in these experiments.

Example 10

In Vitro Analysis: Antisense Inhibition of Human Androgen Receptor mRNA

Measurement of Proliferating Viable Cells (MTS Assay)

LNCaP prostate cancer and A549 lung cancer cells were seeded to a density of 150,000 cells per well in a 6-well plate the day prior to transfection. A549 cells were cultured in DMEM (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 μg/ml) whereas LNCaP cells were cultured in RPMI 1640 Medium (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 μg/ml). On the following day, medium was removed followed by addition of 1.2 ml OptiMEM containing 5 μg/ml Lipofectamine-2000 (Invitrogen). Cells were incubated for 7 min before adding 0.3 ml oligonucleotides diluted in OptiMEM. The final oligonucleotide concentrations were 4 nM and 16 nM. After 4 hours of treatment, media was removed and cells

were trypsinized and seeded to a density of 5000 cells per well in a clear 96 well plate (Scientific Orange no. 1472030100) in 100 µl media. Viable cells were measured at the times indicated by adding 10 µl the tetrazolium compound [3-(4,5-dimethyl-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt; MTS] and an electron coupling reagent (phenazine ethosulfate; PES) (CellTiter 96® AQueous One Solution Cell Proliferation Assay, Promega). Viable cells were measured at 490 nm in a Powerwave (Biotek Instruments). The OD490 nm measurements were plotted against time/h. (See FIG. **13** and FIG. **14**). As shown in FIG. **13** and FIG. **14**, oligonucleotides of SEQ ID NOs: 58 and 77 inhibit growth of both LNCaP prostate and A549 lung cancer cells.

Example 11

In Vitro Analysis: Caspase 3/7 Activity by Antisense Inhibition of Hunan Androgen Receptor mRNA

LNCaP prostate cancer cells and A549 lung cancer cells were seeded to a density of 150,000 cells per well in a 6-well plate the day prior to transfection. A549 cells were cultured in DMEM (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 µg/ml) whereas LNCaP cells were cultured in RPMI 1640 Medium (Sigma)+10% fetal bovine serum (PBS)+2 mM Glutamax I+gentamicin (25 µg/ml). The next day medium was removed followed by addition of 1.2 ml OptiMEM containing 5 µg/ml Lipofectamine2000 (Invitrogen). Cells were incubated for 7 min before adding 0.3 ml oligonucleotides diluted in OptiMEM. The final oligonucleotide concentrations were 4 nM and 16 nM. After 4 hours of treatment, media was removed and cells were trypsinized and seeded to a density of 5000 cells per well in a white 96 well plate (Nunc) in 100 µl media. Caspase 3/7 activity was measured at the times indicated by adding 100 µl Caspase-Glo 3/7 assay (Promega). Caspase 3/7 activity was measured using a luminometer. The caspase 3/7 activities were measured at three different time points 14 h, 48 h and 72 h (See FIG. **15** and FIG. **16**). As shown in FIG. **15** and FIG. **16**, oligonucleotides of SEQ ID NOs: 58 and 77 induce caspase 3/7 activity in both LNCaP prostate and A549 lung cancer cells.

Example 12

In Vitro Analysis: Antisense Inhibition of Human Androgen Receptor mRNA Expression by Oligonucleotide Compounds in Prostate Cancer Cell Line LNCaP and Lung Cancer Cell Line A549

Oligonucleotides were evaluated for their potential to knock down androgen receptor mRNA expression at concentrations of 0.5, 1, 2, 4, 8 and 16 nM (see FIG. **11**). LNCaP prostate cancer cells and A549 lung cancer cells were seeded to a density of 150,000 cells per well in a 6-well plate the day prior to transfection. A549 cells were cultured in DMEM (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 µg/ml). LNCaP cells were cultured in RPMI 1640 Medium (Sigma)+10% fetal bovine serum (FBS)+2 mM Glutamax I+gentamicin (25 µg/ml). On the following day, medium was removed followed by addition of 1.2 ml OptiMEM containing 5 µg/ml Lipofectamine2000 (Invitrogen). Cells were incubated for 7 min before adding 0.3 ml oligonucleotides diluted in OptiMEM. The final oligonucleotide concentrations were 0.5, 1, 2, 4, 8 and 16 nM. Cells were washed and serum-containing media was added. After oligomer treatment cells were allowed to recover for 20 hours before they were harvested for RNA analysis. The procedure for RNA isolation, cDNA synthesis and qPCR were as described in Examples 5, 6 and 7. As shown in FIGS. **11** and **12** oligonucleotides of SEQ ID NOs: 58 and 77 were potent in knocking down AR mRNA expression in both the lung cancer cell line A549 and in the androgen receptor-dependent LNCaP prostate cancer cell line.

Example 13

In Vivo Analysis: Effect of Antisense Oligonucleotides on PSA Levels and Androgen-Dependent Prostate Tumor Growth in Mice

Six to seven week old male athymic nu/nu mice (Harlan Sprague Dawley) weighing an average of 27.3±2.4 g were used in the study. Ten million cells of 22RV1 (androgen-independent prostate cancer line) were suspended in PBS (Gibco#14190) and Matrigel (BD#356234) with a ratio of 1:1 were injected subcutaneously into each mouse. When tumors reached an average volume of 150-200 mm^3, the mice were divided into nine experimental groups. Two hundred µl of oligomer were injected intravenously when the average tumor size reached 152.66±27.97 mm^3. Oligomers were given every 3 days for a total of 5 dosings. The control vehicles were given using the same dosing regimen as the oligomers. On day 16, mice were sacrificed and blood collected in EDTA laced tubes and spun for 5 min. 50 µl of the supernatants were then subjected to PSA assay using the ELISA kit from ALPCO Diagnostics in Salem (PSAHU-L01). Results of the experiment are shown in FIG. **17**.

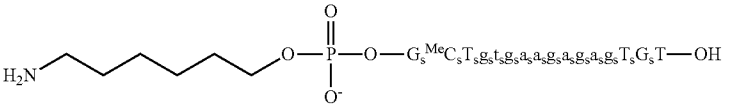
Six to seven week old male athymic nu/nu mice (Harlan Sprague Dawley) weighing an average of 27.3±2.4 g were used in the study. Ten million cells of 22RV1 (androgen-independent prostate cancer line) were suspended in PBS (Gibco#14190) and Matrigel (BD#356234) with a ratio of 1:1 were injected subcutaneously into each mouse. When tumors reached an average volume of 150-200 mm^3, the mice were divided into nine experimental groups. Two hundred µl of oligomer was injected intravenously when the average tumor size reached 152.66±27.97 mm^3. Oligomers were given every 3 days for a total of 5 dosings. The control vehicles were given using the same dosing regimen as the oligomers. The tumor volumes for each mouse were determined by measuring two dimensions with calipers and calculated using the formula: tumor volume=(length×width^2)/2). Results of the experiment are shown in FIG. **18**.

Example 14

Preparation of Conjugates of Oligomers with Polyethylene Glycol

The oligomers having sequences shown as SEQ ID NO: 48 or SEQ ID NO: 63 are functionalized on the 5' terminus by attaching an aminoalkyl group, such as hexan-1-amine blocked with a blocking group such as Fmoc to the 5' phosphate groups of the oligomers using routine phosphoramidite chemistry, oxidizing the resultant compounds, deprotecting them and purifying them to achieve the functionalized oligomers, respectively, having the formulas (IA) and (IB):

(IA)



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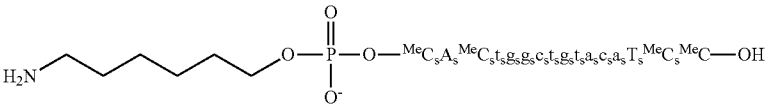
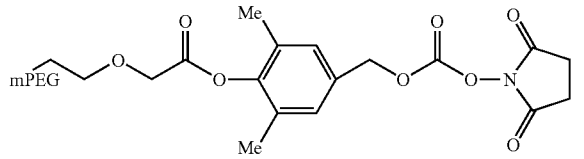


wherein the bold uppercase letters represent nucleoside analogue monomers, lowercase letters represent DNA monomers, the subscript "s" represents a phosphorothioate linkage, and ^{Me}C represents 5-methylcytosine.

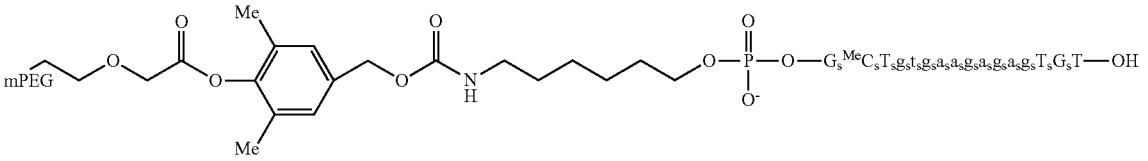
A solution of activated PEG, such as the one shown in formula (II):

(II)

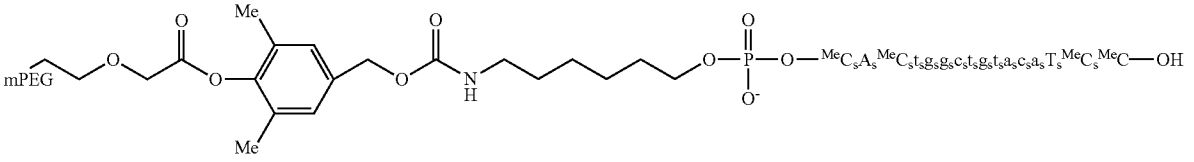


wherein the PEG moiety has an average molecular weight of 12,000, and each of the compounds of formulas (IA) and (IB) in PBS buffer are stirred in separate vessels at room temperature for 12 hours. The reaction solutions are extracted three times with methylene chloride and the combined organic layers are dried over magnesium sulphate and filtered and the solvent is evaporated under reduced pressure. The resulting residues are dissolved in double distilled water and loaded onto an anion exchange column. Unreacted PEG linker is eluted with water and the products are eluted with NH_4HCO_3 solution. Fractions containing pure products are pooled and lypophilized to yield the conjugates SEQ ID NOs: 48 and 63, respectively as show in formulas (IIIA) and (IIIB):

(IIIA)



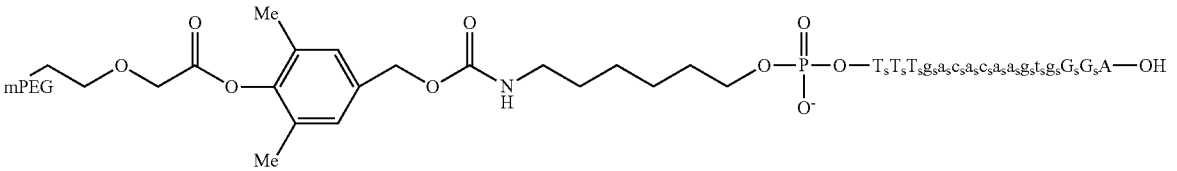
(IIIB)



wherein each of the oligomers of SEQ ID NOs: 48 and 63 is attached to a PEG polymer having average molecular weight of 12,000 via a releasable linker.

Chemical structures of PEG polymer conjugates that can be made with oligomers having sequences shown in SEQ NOs: 51, 58 and 77 using the process described above are respectively shown in formulas (IVA), (IVB) and (IVC):

(IVA)



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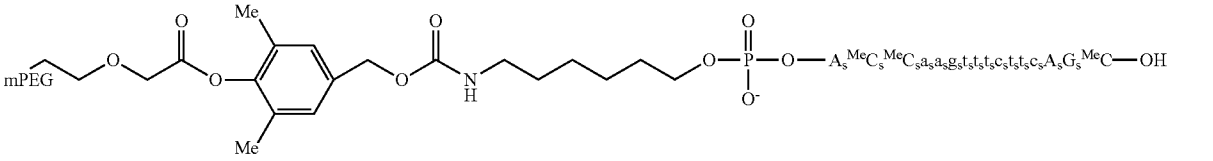
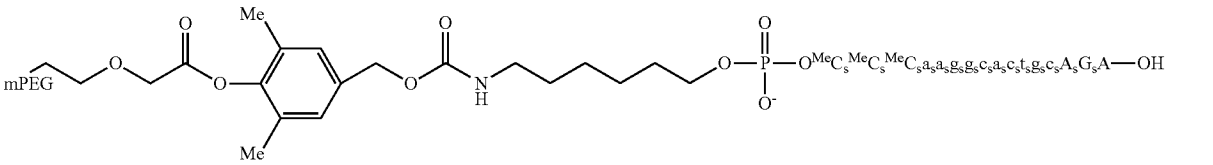
(IVB)



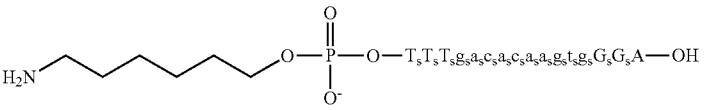
(IVC)



wherein bold uppercase letters represent beta-D-oxy-LNA monomers, lowercase letters represent DNA monomers, the subscript "s" represents a phosphorothioate linkage and ^{Me}C represent 5-methylcytosine.

Activated oligomers that can be used in this process to respectively make the conjugates shown in formulas (NA), (IVB) and (IVC) have the chemical structures shown in formulas (VA), (VB) and (VC):

(VA)



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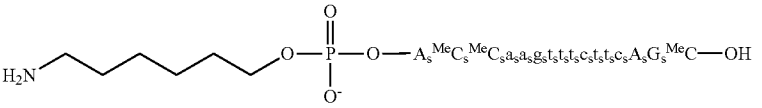
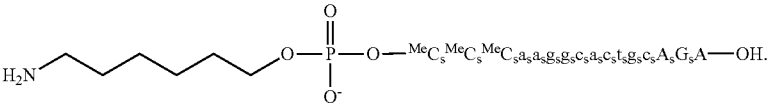


(VC)



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phosphorothioate

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gtgacaccca gaagct 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 9

catccctgct tcataa 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 10

accaagtttc ttcagc 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 11

cttggcccac ttgacc 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

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tcctggagtt gacatt 16

<210> SEQ ID NO 13
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<221> NAME/KEY: misc_feature
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

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cactggctgt acatcc 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 14

catccaaact cttgag 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 15

gctttcatgc acagga 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 16

gaagttcatc aaagaa 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

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agttccttga tgtagt 16

<210> SEQ ID NO 18
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ttgcacagag atgatc 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 19

gatgggcttg actttc 16

<210> SEQ ID NO 20
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caggcagaag acatct 16

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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 21

cccaaggcac tgcaga 16

<210> SEQ ID NO 22
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 22

gctgacattc atagcc 16

<210> SEQ ID NO 23
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<221> NAME/KEY: misc_feature
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<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 23

gagaaccatc ctcacc 16

<210> SEQ ID NO 24
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 24

ggaccaggta gcctgt 16

<210> SEQ ID NO 25
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<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 25

cccctggact cagatg 16

<210> SEQ ID NO 26
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 26

gcacaaggag tgggac 16

<210> SEQ ID NO 27
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 27

gctgtgaaga gagtgt 16

<210> SEQ ID NO 28
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 28

tttgacacaa gtggga 16

<210> SEQ ID NO 29
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<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 29

gtgacaccca gaagct 16

<210> SEQ ID NO 30
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<212> TYPE: DNA
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
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<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 30

catccctgct tcataa 16

<210> SEQ ID NO 31
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<212> TYPE: DNA
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<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 31

accaagtttc ttcagc 16

<210> SEQ ID NO 32
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 32

cttggcccac ttgacc 16

<210> SEQ ID NO 33
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<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 33

tcctggagtt gacatt 16

<210> SEQ ID NO 34
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<212> TYPE: DNA
<213> ORGANISM: artificial
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 34

cactggctgt acatcc 16

<210> SEQ ID NO 35
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<212> TYPE: DNA
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 35

catccaaact cttgag 16

<210> SEQ ID NO 36
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<212> TYPE: DNA
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 36

gctttcatgc acagga 16

<210> SEQ ID NO 37
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<212> TYPE: DNA
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 37

gaagttcatc aaagaa 16

<210> SEQ ID NO 38
<211> LENGTH: 16
<212> TYPE: DNA
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 38

agttccttga tgtagt 16

<210> SEQ ID NO 39
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<212> TYPE: DNA
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 39

ttgcacagag atgatc 16

<210> SEQ ID NO 40
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<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 40

gatgggcttg actttc 16

<210> SEQ ID NO 41
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 41

caggcagaag acatct 16

<210> SEQ ID NO 42
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<212> TYPE: DNA
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<220> FEATURE:
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<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 42

cccaaggcac tgcaga 16

<210> SEQ ID NO 43
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 Gapmer - optionally phosphorothioate

<400> SEQUENCE: 43

gctgacattc atagcc 16

<210> SEQ ID NO 44
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<212> TYPE: DNA
<213> ORGANISM: artificial
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 44

gagaaccatc ctcacc 16

<210> SEQ ID NO 45
<211> LENGTH: 16
<212> TYPE: DNA
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 45

ggaccaggta gcctgt 16

<210> SEQ ID NO 46
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<212> TYPE: DNA
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<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 46

cccctggact cagatg 16

<210> SEQ ID NO 47
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 47

gcacaaggag tgggac 16

<210> SEQ ID NO 48
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<212> TYPE: DNA
<213> ORGANISM: artificial
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 48

gctgtgaaga gagtgt 16

<210> SEQ ID NO 49
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<212> TYPE: DNA
<213> ORGANISM: artificial
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<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 49

ctgtgaagag agtg 14

<210> SEQ ID NO 50
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 50

tgtgaagaga gt 12

<210> SEQ ID NO 51
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 51

tttgacacaa gtggga 16

<210> SEQ ID NO 52
<211> LENGTH: 14
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 52

ttgacacaag tggg 14

<210> SEQ ID NO 53
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:

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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 53

tgacacaagt gg 12

<210> SEQ ID NO 54
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 54

gtgacaccca gaagct 16

<210> SEQ ID NO 55
<211> LENGTH: 14
<212> TYPE: DNA
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<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 55

tgacacccag aagc 14

<210> SEQ ID NO 56
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 56

gacacccaga ag 12

<210> SEQ ID NO 57
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 57

catccctgct tcataa 16

<210> SEQ ID NO 58
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 58

accaagtttc ttcagc 16

<210> SEQ ID NO 59
<211> LENGTH: 14
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 59

ccaagtttct tcag 14

<210> SEQ ID NO 60
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 60

caagtttctt ca 12

<210> SEQ ID NO 61
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 61

cttggcccac ttgacc 16

<210> SEQ ID NO 62
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 62

tcctggagtt gacatt 16

<210> SEQ ID NO 63
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 63

cactggctgt acatcc 16

<210> SEQ ID NO 64
<211> LENGTH: 14
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 64

actggctgta catc 14

<210> SEQ ID NO 65
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 65

ctggctgtac at 12

<210> SEQ ID NO 66
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 66

catccaaact cttgag 16

<210> SEQ ID NO 67
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 67

gctttcatgc acagga 16

<210> SEQ ID NO 68
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:

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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 68

gaagttcatc aaagaa 16

<210> SEQ ID NO 69
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 69

agttccttga tgtagt 16

<210> SEQ ID NO 70
<211> LENGTH: 14
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 70

gttccttgat gtag 14

<210> SEQ ID NO 71
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 71

ttccttgatg ta 12

<210> SEQ ID NO 72
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 72

ttgcacagag atgatc 16

<210> SEQ ID NO 73
<211> LENGTH: 16
<212> TYPE: DNA

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<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 73

gatgggcttg actttc 16

<210> SEQ ID NO 74
<211> LENGTH: 14
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 74

atgggcttga cttt 14

<210> SEQ ID NO 75
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 75

tgggcttgac tt 12

<210> SEQ ID NO 76
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 76

caggcagaag acatct 16

<210> SEQ ID NO 77
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 77

cccaaggcac tgcaga 16

<210> SEQ ID NO 78
<211> LENGTH: 14
<212> TYPE: DNA

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<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(14)
<223> OTHER INFORMATION: 3-9-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 78

ccaaggcact gcag 14

<210> SEQ ID NO 79
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(12)
<223> OTHER INFORMATION: 2-8-2 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 79

caaggcactg ca 12

<210> SEQ ID NO 80
<211> LENGTH: 16
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(16)
<223> OTHER INFORMATION: 3-10-3 LNA Gapmer - fully phosphorothioate

<400> SEQUENCE: 80

gctgacattc atagcc 16

<210> SEQ ID NO 81
<211> LENGTH: 2999
<212> TYPE: DNA
<213> ORGANISM: Mus musculus

<400> SEQUENCE: 81

gaattcggtg gaagctacag acaagctcaa ggatggaggt gcagttaggg ctgggaaggg 60

tctacccacg gcccccatcc aagacctatc gaggagcgtt ccagaatctg ttccagagcg 120

tgcgcgaagc gatccagaac ccgggcccca ggcaccctga ggccgctaac atagcacctc 180

ccggcgcctg tttacagcag aggcaggaga ctagcccccg gcggcggcgg cggcagcagc 240

acactgagga tggttctcct caagcccaca tcagaggccc cacaggctac ctggccctgg 300

aggaggaaca gcagccttca cagcagcagg cagcctccga gggccaccct gagagcagct 360

gcctccccga gcctggggcg gccaccgctc ctggcaaggg gctgccgcag cagccaccag 420

ctcctccaga tcaggatgac tcagctgccc catccacgtt gtccctgctg ggccccactt 480

tcccaggctt aagcagctgc tccgccgaca ttaaagacat tttgaacgag gccggcacca 540

tgcaacttct tcagcagcag caacaacagc agcagcacca acagcagcac caacagcacc 600

aacagcagca ggaggtaatc tccgaaggca gcagcgcaag agccagggag gccacggggg 660

ctccctcttc ctccaaggat agttacctag ggggcaattc aaccatatct gacagtgcca 720

aggagttgtg taaagcagtg tctgtgtcca tgggattggg tgtggaagca ttggaacatc 780

tgagtccagg ggaacagctt cggggagact gcatgtacgc gtcgctcctg ggaggtccac 840

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ccgcggtgcg tcccactcct tgtgcgccgc tgcccgaatg caaaggtctt cccctggacg 900

aaggcccagg caaaagcact gaagagactg ctgagtattc ctctttcaag ggaggttacg 960

ccaaaggatt ggaaggtgag agcttggggt gctctggcag cagtgaagca ggtagctctg 1020

ggacacttga gatcccgtcc tctctgtctc tgtataaatc tggagcacta gacgaggcag 1080

cagcatacca gaatcgcgac tactacaact ttccgctggc tctgtccggg ccgccgcacc 1140

ccccgccccc tacccatcca cacgcccgta tcaagctgga gaacccattg gactacggca 1200

gcgcctgggc tgcggcggca gcgcaatgcc gctatgggga cttgggtagt ctacatggag 1260

ggagtgtagc cgggcccagc actggatcgc ccccagccac cacctcttct tcctggcata 1320

ctctcttcac agctgaagaa ggccaattat atgggccagg aggcgggggc ggcagcagca 1380

gcccaagcga tgccgggcct gtagcccccc atggctacac tcggcccccc caggggctga 1440

caagccagga gagtgactac tctgcctccg aagtgtggta tcctggtgga gttgtgaaca 1500

gagtacccta tcccagtccc aattgtgtca aaagtgaaat gggaccttgg atggagaact 1560

actccggacc ttatggggac atgcgtttgg acagtaccag ggaccatgtt ttacccatcg 1620

actattactt tccaccccag aagacctgcc tgatctgtgg agatgaagct tctggctgtc 1680

actacggagc tctcacttgt ggcagctgca aggtcttctt caaaagagcc gctgaaggga 1740

aacagaagta tctatgtgcc agcagaaacg attgtaccat tgataaattt cggaggaaaa 1800

attgcccatc ttgtcgtctc cggaaatgtt atgaagcagg gatgactctg ggagctcgta 1860

agctgaagaa acttggaaat ctaaaactac aggaggaagg agaaaactcc aatgctggca 1920

gccccactga ggacccatcc cagaagatga ctgtatcaca cattgaaggc tatgaatgtc 1980

agcctatctt tcttaacgtc ctggaagcca ttgagccagg agtggtgtgt gccggacatg 2040

acaacaacca accagattcc tttgctgcct tgttatctag cctcaatgag cttggagaga 2100

ggcagcttgt gcatgtggtc aagtgggcca aggccttgcc tggcttccgc aacttgcatg 2160

tggatgacca gatggcggtc attcagtatt cctggatggg actgatggta tttgccatgg 2220

gttggcggtc cttcactaat gtcaactcca ggatgctcta ctttgcacct gacttggttt 2280

tcaatgagta ccgcatgcac aagtctcgga tgtacagcca gtgtgtgagg atgaggcacc 2340

tgtctcaaga gtttggatgg ctccaaataa ccccccagga attcctgtgc atgaaagcac 2400

tgctgctctt cagcattatt ccagtggatg ggctgaaaaa tcaaaaattc tttgatgaac 2460

ttcgaatgaa ctacatcaag gaactcgatc gcatcattgc atgcaaaaga aagaatccca 2520

catcctgctc aaggcgcttc taccagctca ccaagctcct ggattctgtg cagcctattg 2580

caagagagct gcatcagttc acttttgacc tgctaatcaa gtcccatatg gtgagcgtgg 2640

actttcctga aatgatggca gagatcatct ctgtgcaagt gcccaagatc ctttctggga 2700

aagtcaagcc catctatttc cacacacagt gaagatttgg aaaccctaat acccaaaacc 2760

caccttgttc cctttccaga tgtcttctgc ctgttatata actctgcact acttctctgc 2820

agtgccttgg gggaaattcc tctactgatg tacagtctgt cgtgaacagg ttcctcagtt 2880

ctatttcctg ggcttctcct tctttttttt tcttcttccc tccctctttc accctcccat 2940

ggcacatttt gaatctgctg cgtattgtgg ctcctgcctt tgttttgatt tctgttgta 2999

<210> SEQ ID NO 82
<211> LENGTH: 3175
<212> TYPE: DNA
<213> ORGANISM: Macaca mulatta

<400> SEQUENCE: 82

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cccaaaaaat aaaaacaaac aaaaacaaaa caaaacaaaa aaaacgaata aagaaaaagg     60

taataactca gttcttattt gcacctactt ccagtggaca ctgaatttgg aaggtggagg    120

attcttgttt tttcttttaa gatcgggcat cttttgaatc tacccctcaa gtgttaagag    180

acagactgtg agcctagcag ggcagatctt gtccaccgtg tgtcttcttt tgcaggagac    240

tttgaggctg tcagagcgct ttttgcgtgg ttgctcccgc aagtttcctt ctctggagct    300

tcccgcaggt gggcagctag ctgcagcgac taccgcatca tcacagcctg ttgaactctt    360

ctgagcaaga gaaggggagg cggggtaagg gaagtaggtg gaagattcag ccaagctcaa    420

ggatggaggt gcagttaggg ctggggaggg tctaccctcg gccgccgtcc aagacctacc    480

gaggagcttt ccagaatctg ttccagagcg tgcgcgaagt gatccagaac ccgggcccca    540

ggcacccaga ggccgcgagc gcagcacctc ccggcgccag tttgcagcag cagcagcagc    600

agcagcaaga aactagcccc cggcaacagc agcagcagca gcagggtgag gatggttctc    660

cccaagccca tcgtagaggc cccacaggct acctggtcct ggatgaggaa cagcagcctt    720

cacagcctca gtcagccccg gagtgccacc ccgagagagg ttgcgtccca gagcctggag    780

ccgccgtggc cgccggcaag gggctgccgc agcagctgcc agcacctccg gacgaggatg    840

actcagctgc cccatccacg ttgtctctgc tgggccccac tttccccggc ttaagcagct    900

gctccgccga ccttaaagac atcctgagcg aggccagcac catgcaactc cttcagcaac    960

agcagcagga agcagtatcc gaaggcagca gcagcgggag agcgagggag gcctcggggg   1020

ctcccacttc ctccaaggac aattacttag agggcacttc gaccatttct gacagcgcca   1080

aggagctgtg taaggcagtg tcggtgtcca tgggcttggg tgtggaggcg ttggagcatc   1140

tgagtccagg ggaacagctt cgggggggatt gcatgtacgc cccagttttg ggagttccac   1200

ccgctgtgcg tcccactccg tgtgccccat tggccgaatg caaaggttct ctgctagacg   1260

acagcgcagg caagagcact gaagatactg ctgagtattc ccctttcaag ggaggttaca   1320

ccaaagggct agaaggcgag agcctaggct gctctggcag cgctgcagca gggagctccg   1380

ggacacttga actgccgtcc accctgtctc tctacaagtc cggagcactg gacgaggcag   1440

ctgcgtacca gagtcgcgac tactacaact ttccactggc tctggccggg ccgccgcccc   1500

ctccaccgcc tccccatccc cacgctcgca tcaagctgga gaacccgctg gactatggca   1560

gcgcctgggc ggctgcggcg gcgcagtgcc gctatgggga cctggcgagc ctgcatggcg   1620

cgggtgcagc gggacccggc tctgggtcac cctcagcggc cgcttcctca tcctggcaca   1680

ctctcttcac agccgaagaa ggccagttgt atggaccgtg tggtggtggg ggcggcggcg   1740

gtggcggcgg cggcggcggc gcaggcgagg cgggagctgt agccccctac ggctacactc   1800

ggccacctca ggggctggcg ggccaggaag gcgacttcac cgcacctgat gtgtggtacc   1860

ctggcggcat ggtgagcaga gtgccctatc ccagtcccac ttgtgtcaaa agcgagatgg   1920

gcccctggat ggatagctac tccggacctt acggggacat gcgtttggag actgccaggg   1980

accatgtttt gccaattgac tattactttc caccccagaa gacctgcctg atctgtggag   2040

atgaagcttc tgggtgtcac tatggagctc tcacatgtgg aagctgcaag gtcttcttca   2100

aaagagccgc tgaagggaaa cagaagtacc tgtgtgccag cagaaatgat tgcactattg   2160

ataaattccg aaggaaaaat tgtccatctt gccgtcttcg gaaatgttat gaagcaggga   2220

tgactctggg agcccggaag ctgaagaaac ttggtaatct gaaactacag gaggaaggag   2280

aggcttccag caccaccagc cccactgagg agacagccca gaagctgaca gtgtcacaca   2340

ttgaaggcta tgaatgtcag cccatctttc tgaatgtcct ggaggccatt gagccaggtg   2400
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tggtgtgtgc tggacatgac aacaaccagc ccgactcctt cgcagccttg ctctctagcc 2460

tcaatgaact gggagagaga cagcttgtac atgtggtcaa gtgggccaag gccttgcctg 2520

gcttccgcaa cttacacgtg gacgaccaga tggctgtcat tcagtactcc tggatggggc 2580

tcatggtgtt tgccatgggc tggcgatcct tcaccaatgt caactccagg atgctctact 2640

ttgcccctga tctggttttc aatgagtacc gcatgcacaa atcccggatg tacagccagt 2700

gtgtccgaat gaggcacctc tctcaagagt ttggatggct ccaaatcacc ccccaggaat 2760

tcctgtgcat gaaagcgctg ctactcttca gcattattcc agtggatggg ctgaaaaatc 2820

aaaaattctt tgatgaactt cgaatgaact acatcaagga actcgatcgt atcattgcat 2880

gcaaaagaaa aaatcccaca tcctgctcaa ggcgtttcta ccagctcacc aagctcctgg 2940

actccgtgca gcctattgcg agagagctgc atcagttcac ttttgacctg ctaatcaagt 3000

cacacatggt gagcgtggac tttccggaaa tgatggcaga gatcatctct gtgcaagtgc 3060

ccaagatcct ttctgggaaa gtcaagccca tctatttcca cacccagtga agcattggaa 3120

atccctattt cctcacccca gctcatgccc cctttcagat gtcttctgcc tgtta 3175

<210> SEQ ID NO 83
<211> LENGTH: 920
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 83

Met Glu Val Gln Leu Gly Leu Gly Arg Val Tyr Pro Arg Pro Pro Ser
1 5 10 15

Lys Thr Tyr Arg Gly Ala Phe Gln Asn Leu Phe Gln Ser Val Arg Glu
20 25 30

Val Ile Gln Asn Pro Gly Pro Arg His Pro Glu Ala Ala Ser Ala Ala
35 40 45

Pro Pro Gly Ala Ser Leu Leu Leu Leu Gln Gln Gln Gln Gln Gln Gln
50 55 60

Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln
65 70 75 80

Glu Thr Ser Pro Arg Gln Gln Gln Gln Gln Gly Glu Asp Gly Ser
85 90 95

Pro Gln Ala His Arg Arg Gly Pro Thr Gly Tyr Leu Val Leu Asp Glu
100 105 110

Glu Gln Gln Pro Ser Gln Pro Gln Ser Ala Leu Glu Cys His Pro Glu
115 120 125

Arg Gly Cys Val Pro Glu Pro Gly Ala Ala Val Ala Ala Ser Lys Gly
130 135 140

Leu Pro Gln Gln Leu Pro Ala Pro Pro Asp Glu Asp Asp Ser Ala Ala
145 150 155 160

Pro Ser Thr Leu Ser Leu Leu Gly Pro Thr Phe Pro Gly Leu Ser Ser
165 170 175

Cys Ser Ala Asp Leu Lys Asp Ile Leu Ser Glu Ala Ser Thr Met Gln
180 185 190

Leu Leu Gln Gln Gln Gln Gln Glu Ala Val Ser Glu Gly Ser Ser Ser
195 200 205

Gly Arg Ala Arg Glu Ala Ser Gly Ala Pro Thr Ser Ser Lys Asp Asn
210 215 220

Tyr Leu Gly Gly Thr Ser Thr Ile Ser Asp Asn Ala Lys Glu Leu Cys
225 230 235 240

Lys Ala Val Ser Val Ser Met Gly Leu Gly Val Glu Ala Leu Glu His

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               245                 250                 255

Leu Ser Pro Gly Glu Gln Leu Arg Gly Asp Cys Met Tyr Ala Pro Leu
            260                 265                 270

Leu Gly Val Pro Pro Ala Val Arg Pro Thr Pro Cys Ala Pro Leu Ala
        275                 280                 285

Glu Cys Lys Gly Ser Leu Leu Asp Asp Ser Ala Gly Lys Ser Thr Glu
    290                 295                 300

Asp Thr Ala Glu Tyr Ser Pro Phe Lys Gly Gly Tyr Thr Lys Gly Leu
305                 310                 315                 320

Glu Gly Glu Ser Leu Gly Cys Ser Gly Ser Ala Ala Ala Gly Ser Ser
                325                 330                 335

Gly Thr Leu Glu Leu Pro Ser Thr Leu Ser Leu Tyr Lys Ser Gly Ala
            340                 345                 350

Leu Asp Glu Ala Ala Ala Tyr Gln Ser Arg Asp Tyr Tyr Asn Phe Pro
        355                 360                 365

Leu Ala Leu Ala Gly Pro Pro Pro Pro Pro Pro Pro Pro His Pro His
    370                 375                 380

Ala Arg Ile Lys Leu Glu Asn Pro Leu Asp Tyr Gly Ser Ala Trp Ala
385                 390                 395                 400

Ala Ala Ala Ala Gln Cys Arg Tyr Gly Asp Leu Ala Ser Leu His Gly
                405                 410                 415

Ala Gly Ala Ala Gly Pro Gly Ser Gly Ser Pro Ser Ala Ala Ala Ser
            420                 425                 430

Ser Ser Trp His Thr Leu Phe Thr Ala Glu Glu Gly Gln Leu Tyr Gly
        435                 440                 445

Pro Cys Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly
    450                 455                 460

Gly Gly Gly Gly Gly Gly Gly Gly Gly Glu Ala Gly Ala Val Ala Pro
465                 470                 475                 480

Tyr Gly Tyr Thr Arg Pro Pro Gln Gly Leu Ala Gly Gln Glu Ser Asp
                485                 490                 495

Phe Thr Ala Pro Asp Val Trp Tyr Pro Gly Gly Met Val Ser Arg Val
            500                 505                 510

Pro Tyr Pro Ser Pro Thr Cys Val Lys Ser Glu Met Gly Pro Trp Met
        515                 520                 525

Asp Ser Tyr Ser Gly Pro Tyr Gly Asp Met Arg Leu Glu Thr Ala Arg
    530                 535                 540

Asp His Val Leu Pro Ile Asp Tyr Tyr Phe Pro Pro Gln Lys Thr Cys
545                 550                 555                 560

Leu Ile Cys Gly Asp Glu Ala Ser Gly Cys His Tyr Gly Ala Leu Thr
                565                 570                 575

Cys Gly Ser Cys Lys Val Phe Phe Lys Arg Ala Ala Glu Gly Lys Gln
            580                 585                 590

Lys Tyr Leu Cys Ala Ser Arg Asn Asp Cys Thr Ile Asp Lys Phe Arg
        595                 600                 605

Arg Lys Asn Cys Pro Ser Cys Arg Leu Arg Lys Cys Tyr Glu Ala Gly
    610                 615                 620

Met Thr Leu Gly Ala Arg Lys Leu Lys Lys Leu Gly Asn Leu Lys Leu
625                 630                 635                 640

Gln Glu Glu Gly Glu Ala Ser Ser Thr Thr Ser Pro Thr Glu Glu Thr
                645                 650                 655

Thr Gln Lys Leu Thr Val Ser His Ile Glu Gly Tyr Glu Cys Gln Pro
            660                 665                 670
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Ile Phe Leu Asn Val Leu Glu Ala Ile Glu Pro Gly Val Val Cys Ala
        675                 680                 685

Gly His Asp Asn Asn Gln Pro Asp Ser Phe Ala Ala Leu Leu Ser Ser
    690                 695                 700

Leu Asn Glu Leu Gly Glu Arg Gln Leu Val His Val Val Lys Trp Ala
705                 710                 715                 720

Lys Ala Leu Pro Gly Phe Arg Asn Leu His Val Asp Asp Gln Met Ala
                725                 730                 735

Val Ile Gln Tyr Ser Trp Met Gly Leu Met Val Phe Ala Met Gly Trp
            740                 745                 750

Arg Ser Phe Thr Asn Val Asn Ser Arg Met Leu Tyr Phe Ala Pro Asp
        755                 760                 765

Leu Val Phe Asn Glu Tyr Arg Met His Lys Ser Arg Met Tyr Ser Gln
    770                 775                 780

Cys Val Arg Met Arg His Leu Ser Gln Glu Phe Gly Trp Leu Gln Ile
785                 790                 795                 800

Thr Pro Gln Glu Phe Leu Cys Met Lys Ala Leu Leu Leu Phe Ser Ile
                805                 810                 815

Ile Pro Val Asp Gly Leu Lys Asn Gln Lys Phe Phe Asp Glu Leu Arg
            820                 825                 830

Met Asn Tyr Ile Lys Glu Leu Asp Arg Ile Ile Ala Cys Lys Arg Lys
        835                 840                 845

Asn Pro Thr Ser Cys Ser Arg Arg Phe Tyr Gln Leu Thr Lys Leu Leu
    850                 855                 860

Asp Ser Val Gln Pro Ile Ala Arg Glu Leu His Gln Phe Thr Phe Asp
865                 870                 875                 880

Leu Leu Ile Lys Ser His Met Val Ser Val Asp Phe Pro Glu Met Met
                885                 890                 895

Ala Glu Ile Ile Ser Val Gln Val Pro Lys Ile Leu Ser Gly Lys Val
            900                 905                 910

Lys Pro Ile Tyr Phe His Thr Gln
        915                 920


<210> SEQ ID NO 84
<211> LENGTH: 899
<212> TYPE: PRT
<213> ORGANISM: Mus musculus

<400> SEQUENCE: 84

Met Glu Val Gln Leu Gly Leu Gly Arg Val Tyr Pro Arg Pro Pro Ser
1               5                   10                  15

Lys Thr Tyr Arg Gly Ala Phe Gln Asn Leu Phe Gln Ser Val Arg Glu
            20                  25                  30

Ala Ile Gln Asn Pro Gly Pro Arg His Pro Glu Ala Ala Asn Ile Ala
        35                  40                  45

Pro Pro Gly Ala Cys Leu Gln Gln Arg Gln Glu Thr Ser Pro Arg Arg
    50                  55                  60

Arg Arg Arg Gln Gln His Thr Glu Asp Gly Ser Pro Gln Ala His Ile
65                  70                  75                  80

Arg Gly Pro Thr Gly Tyr Leu Ala Leu Glu Glu Glu Gln Gln Pro Ser
                85                  90                  95

Gln Gln Gln Ala Ala Ser Glu Gly His Pro Glu Ser Ser Cys Leu Pro
            100                 105                 110

Glu Pro Gly Ala Ala Thr Ala Pro Gly Lys Gly Leu Pro Gln Gln Pro
        115                 120                 125
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Pro Ala Pro Pro Asp Gln Asp Asp Ser Ala Ala Pro Ser Thr Leu Ser
    130                 135                 140

Leu Leu Gly Pro Thr Phe Pro Gly Leu Ser Ser Cys Ser Ala Asp Ile
145                 150                 155                 160

Lys Asp Ile Leu Asn Glu Ala Gly Thr Met Gln Leu Leu Gln Gln Gln
                165                 170                 175

Gln Gln Gln Gln Gln His Gln Gln Gln His Gln Gln His Gln Gln Gln
            180                 185                 190

Gln Glu Val Ile Ser Glu Gly Ser Ser Ala Arg Ala Arg Glu Ala Thr
        195                 200                 205

Gly Ala Pro Ser Ser Ser Lys Asp Ser Tyr Leu Gly Gly Asn Ser Thr
    210                 215                 220

Ile Ser Asp Ser Ala Lys Glu Leu Cys Lys Ala Val Ser Val Ser Met
225                 230                 235                 240

Gly Leu Gly Val Glu Ala Leu Glu His Leu Ser Pro Gly Glu Gln Leu
                245                 250                 255

Arg Gly Asp Cys Met Tyr Ala Ser Leu Leu Gly Gly Pro Pro Ala Val
            260                 265                 270

Arg Pro Thr Pro Cys Ala Pro Leu Pro Glu Cys Lys Gly Leu Pro Leu
        275                 280                 285

Asp Glu Gly Pro Gly Lys Ser Thr Glu Glu Thr Ala Glu Tyr Ser Ser
    290                 295                 300

Phe Lys Gly Gly Tyr Ala Lys Gly Leu Glu Gly Glu Ser Leu Gly Cys
305                 310                 315                 320

Ser Gly Ser Ser Glu Ala Gly Ser Ser Gly Thr Leu Glu Ile Pro Ser
                325                 330                 335

Ser Leu Ser Leu Tyr Lys Ser Gly Ala Leu Asp Glu Ala Ala Ala Tyr
            340                 345                 350

Gln Asn Arg Asp Tyr Tyr Asn Phe Pro Leu Ala Leu Ser Gly Pro Pro
        355                 360                 365

His Pro Pro Pro Pro Thr His Pro His Ala Arg Ile Lys Leu Glu Asn
    370                 375                 380

Pro Leu Asp Tyr Gly Ser Ala Trp Ala Ala Ala Ala Ala Gln Cys Arg
385                 390                 395                 400

Tyr Gly Asp Leu Gly Ser Leu His Gly Gly Ser Val Ala Gly Pro Ser
                405                 410                 415

Thr Gly Ser Pro Pro Ala Thr Thr Ser Ser Ser Trp His Thr Leu Phe
            420                 425                 430

Thr Ala Glu Glu Gly Gln Leu Tyr Gly Pro Gly Gly Gly Gly Gly Ser
        435                 440                 445

Ser Ser Pro Ser Asp Ala Gly Pro Val Ala Pro Tyr Gly Tyr Thr Arg
    450                 455                 460

Pro Pro Gln Gly Leu Thr Ser Gln Glu Ser Asp Tyr Ser Ala Ser Glu
465                 470                 475                 480

Val Trp Tyr Pro Gly Gly Val Val Asn Arg Val Pro Tyr Pro Ser Pro
                485                 490                 495

Asn Cys Val Lys Ser Glu Met Gly Pro Trp Met Glu Asn Tyr Ser Gly
            500                 505                 510

Pro Tyr Gly Asp Met Arg Leu Asp Ser Thr Arg Asp His Val Leu Pro
        515                 520                 525

Ile Asp Tyr Tyr Phe Pro Pro Gln Lys Thr Cys Leu Ile Cys Gly Asp
    530                 535                 540

Glu Ala Ser Gly Cys His Tyr Gly Ala Leu Thr Cys Gly Ser Cys Lys
545                 550                 555                 560
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Val Phe Phe Lys Arg Ala Ala Glu Gly Lys Gln Lys Tyr Leu Cys Ala
                565                 570                 575

Ser Arg Asn Asp Cys Thr Ile Asp Lys Phe Arg Arg Lys Asn Cys Pro
            580                 585                 590

Ser Cys Arg Leu Arg Lys Cys Tyr Glu Ala Gly Met Thr Leu Gly Ala
        595                 600                 605

Arg Lys Leu Lys Lys Leu Gly Asn Leu Lys Leu Gln Glu Glu Gly Glu
    610                 615                 620

Asn Ser Asn Ala Gly Ser Pro Thr Glu Asp Pro Ser Gln Lys Met Thr
625                 630                 635                 640

Val Ser His Ile Glu Gly Tyr Glu Cys Gln Pro Ile Phe Leu Asn Val
                645                 650                 655

Leu Glu Ala Ile Glu Pro Gly Val Val Cys Ala Gly His Asp Asn Asn
            660                 665                 670

Gln Pro Asp Ser Phe Ala Ala Leu Leu Ser Ser Leu Asn Glu Leu Gly
        675                 680                 685

Glu Arg Gln Leu Val His Val Val Lys Trp Ala Lys Ala Leu Pro Gly
    690                 695                 700

Phe Arg Asn Leu His Val Asp Asp Gln Met Ala Val Ile Gln Tyr Ser
705                 710                 715                 720

Trp Met Gly Leu Met Val Phe Ala Met Gly Trp Arg Ser Phe Thr Asn
                725                 730                 735

Val Asn Ser Arg Met Leu Tyr Phe Ala Pro Asp Leu Val Phe Asn Glu
            740                 745                 750

Tyr Arg Met His Lys Ser Arg Met Tyr Ser Gln Cys Val Arg Met Arg
        755                 760                 765

His Leu Ser Gln Glu Phe Gly Trp Leu Gln Ile Thr Pro Gln Glu Phe
    770                 775                 780

Leu Cys Met Lys Ala Leu Leu Leu Phe Ser Ile Ile Pro Val Asp Gly
785                 790                 795                 800

Leu Lys Asn Gln Lys Phe Phe Asp Glu Leu Arg Met Asn Tyr Ile Lys
                805                 810                 815

Glu Leu Asp Arg Ile Ile Ala Cys Lys Arg Lys Asn Pro Thr Ser Cys
            820                 825                 830

Ser Arg Arg Phe Tyr Gln Leu Thr Lys Leu Leu Asp Ser Val Gln Pro
        835                 840                 845

Ile Ala Arg Glu Leu His Gln Phe Thr Phe Asp Leu Leu Ile Lys Ser
    850                 855                 860

His Met Val Ser Val Asp Phe Pro Glu Met Met Ala Glu Ile Ile Ser
865                 870                 875                 880

Val Gln Val Pro Lys Ile Leu Ser Gly Lys Val Lys Pro Ile Tyr Phe
                885                 890                 895

His Thr Gln


<210> SEQ ID NO 85
<211> LENGTH: 895
<212> TYPE: PRT
<213> ORGANISM: Macaca mulatta

<400> SEQUENCE: 85

Met Glu Val Gln Leu Gly Leu Gly Arg Val Tyr Pro Arg Pro Pro Ser
1               5                   10                  15

Lys Thr Tyr Arg Gly Ala Phe Gln Asn Leu Phe Gln Ser Val Arg Glu
            20                  25                  30
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Val Ile Gln Asn Pro Gly Pro Arg His Pro Glu Ala Ala Ser Ala Ala
        35                  40                  45

Pro Pro Gly Ala Ser Leu Gln Gln Gln Gln Gln Gln Gln Gln Glu Thr
    50                  55                  60

Ser Pro Arg Gln Gln Gln Gln Gln Gln Gln Gly Glu Asp Gly Ser Pro
65                  70                  75                  80

Gln Ala His Arg Arg Gly Pro Thr Gly Tyr Leu Val Leu Asp Glu Glu
                85                  90                  95

Gln Gln Pro Ser Gln Pro Gln Ser Ala Pro Glu Cys His Pro Glu Arg
            100                 105                 110

Gly Cys Val Pro Glu Pro Gly Ala Ala Val Ala Ala Gly Lys Gly Leu
        115                 120                 125

Pro Gln Gln Leu Pro Ala Pro Pro Asp Glu Asp Asp Ser Ala Ala Pro
    130                 135                 140

Ser Thr Leu Ser Leu Leu Gly Pro Thr Phe Pro Gly Leu Ser Ser Cys
145                 150                 155                 160

Ser Ala Asp Leu Lys Asp Ile Leu Ser Glu Ala Ser Thr Met Gln Leu
                165                 170                 175

Leu Gln Gln Gln Gln Gln Glu Ala Val Ser Glu Gly Ser Ser Ser Gly
            180                 185                 190

Arg Ala Arg Glu Ala Ser Gly Ala Pro Thr Ser Ser Lys Asp Asn Tyr
        195                 200                 205

Leu Glu Gly Thr Ser Thr Ile Ser Asp Ser Ala Lys Glu Leu Cys Lys
    210                 215                 220

Ala Val Ser Val Ser Met Gly Leu Gly Val Glu Ala Leu Glu His Leu
225                 230                 235                 240

Ser Pro Gly Glu Gln Leu Arg Gly Asp Cys Met Tyr Ala Pro Val Leu
                245                 250                 255

Gly Val Pro Pro Ala Val Arg Pro Thr Pro Cys Ala Pro Leu Ala Glu
            260                 265                 270

Cys Lys Gly Ser Leu Leu Asp Asp Ser Ala Gly Lys Ser Thr Glu Asp
        275                 280                 285

Thr Ala Glu Tyr Ser Pro Phe Lys Gly Gly Tyr Thr Lys Gly Leu Glu
    290                 295                 300

Gly Glu Ser Leu Gly Cys Ser Gly Ser Ala Ala Ala Gly Ser Ser Gly
305                 310                 315                 320

Thr Leu Glu Leu Pro Ser Thr Leu Ser Leu Tyr Lys Ser Gly Ala Leu
                325                 330                 335

Asp Glu Ala Ala Ala Tyr Gln Ser Arg Asp Tyr Tyr Asn Phe Pro Leu
            340                 345                 350

Ala Leu Ala Gly Pro Pro Pro Pro Pro Pro Pro Pro His Pro His Ala
        355                 360                 365

Arg Ile Lys Leu Glu Asn Pro Leu Asp Tyr Gly Ser Ala Trp Ala Ala
    370                 375                 380

Ala Ala Ala Gln Cys Arg Tyr Gly Asp Leu Ala Ser Leu His Gly Ala
385                 390                 395                 400

Gly Ala Ala Gly Pro Gly Ser Gly Ser Pro Ser Ala Ala Ala Ser Ser
                405                 410                 415

Ser Trp His Thr Leu Phe Thr Ala Glu Glu Gly Gln Leu Tyr Gly Pro
            420                 425                 430

Cys Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Ala Gly
        435                 440                 445

Glu Ala Gly Ala Val Ala Pro Tyr Gly Tyr Thr Arg Pro Pro Gln Gly
    450                 455                 460
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Leu Ala Gly Gln Glu Gly Asp Phe Thr Ala Pro Asp Val Trp Tyr Pro
465                 470                 475                 480

Gly Gly Met Val Ser Arg Val Pro Tyr Pro Ser Pro Thr Cys Val Lys
                485                 490                 495

Ser Glu Met Gly Pro Trp Met Asp Ser Tyr Ser Gly Pro Tyr Gly Asp
            500                 505                 510

Met Arg Leu Glu Thr Ala Arg Asp His Val Leu Pro Ile Asp Tyr Tyr
        515                 520                 525

Phe Pro Pro Gln Lys Thr Cys Leu Ile Cys Gly Asp Glu Ala Ser Gly
    530                 535                 540

Cys His Tyr Gly Ala Leu Thr Cys Gly Ser Cys Lys Val Phe Phe Lys
545                 550                 555                 560

Arg Ala Ala Glu Gly Lys Gln Lys Tyr Leu Cys Ala Ser Arg Asn Asp
                565                 570                 575

Cys Thr Ile Asp Lys Phe Arg Arg Lys Asn Cys Pro Ser Cys Arg Leu
            580                 585                 590

Arg Lys Cys Tyr Glu Ala Gly Met Thr Leu Gly Ala Arg Lys Leu Lys
        595                 600                 605

Lys Leu Gly Asn Leu Lys Leu Gln Glu Glu Gly Glu Ala Ser Ser Thr
    610                 615                 620

Thr Ser Pro Thr Glu Glu Thr Ala Gln Lys Leu Thr Val Ser His Ile
625                 630                 635                 640

Glu Gly Tyr Glu Cys Gln Pro Ile Phe Leu Asn Val Leu Glu Ala Ile
                645                 650                 655

Glu Pro Gly Val Val Cys Ala Gly His Asp Asn Asn Gln Pro Asp Ser
            660                 665                 670

Phe Ala Ala Leu Leu Ser Ser Leu Asn Glu Leu Gly Glu Arg Gln Leu
        675                 680                 685

Val His Val Val Lys Trp Ala Lys Ala Leu Pro Gly Phe Arg Asn Leu
    690                 695                 700

His Val Asp Asp Gln Met Ala Val Ile Gln Tyr Ser Trp Met Gly Leu
705                 710                 715                 720

Met Val Phe Ala Met Gly Trp Arg Ser Phe Thr Asn Val Asn Ser Arg
                725                 730                 735

Met Leu Tyr Phe Ala Pro Asp Leu Val Phe Asn Glu Tyr Arg Met His
            740                 745                 750

Lys Ser Arg Met Tyr Ser Gln Cys Val Arg Met Arg His Leu Ser Gln
        755                 760                 765

Glu Phe Gly Trp Leu Gln Ile Thr Pro Gln Glu Phe Leu Cys Met Lys
    770                 775                 780

Ala Leu Leu Leu Phe Ser Ile Ile Pro Val Asp Gly Leu Lys Asn Gln
785                 790                 795                 800

Lys Phe Phe Asp Glu Leu Arg Met Asn Tyr Ile Lys Glu Leu Asp Arg
                805                 810                 815

Ile Ile Ala Cys Lys Arg Lys Asn Pro Thr Ser Cys Ser Arg Arg Phe
            820                 825                 830

Tyr Gln Leu Thr Lys Leu Leu Asp Ser Val Gln Pro Ile Ala Arg Glu
        835                 840                 845

Leu His Gln Phe Thr Phe Asp Leu Leu Ile Lys Ser His Met Val Ser
    850                 855                 860

Val Asp Phe Pro Glu Met Met Ala Glu Ile Ile Ser Val Gln Val Pro
865                 870                 875                 880

Lys Ile Leu Ser Gly Lys Val Lys Pro Ile Tyr Phe His Thr Gln
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885 890 895

<210> SEQ ID NO 86
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 86

tggggagaac catcctcacc ctgc 24

<210> SEQ ID NO 87
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
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<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 87

tccaggacca ggtagcctgt gggg 24

<210> SEQ ID NO 88
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 88

tgttcccctg gactcagatg ctcc 24

<210> SEQ ID NO 89
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 89

tggggcacaa ggagtgggac gcac 24

<210> SEQ ID NO 90
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
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<220> FEATURE:
<221> NAME/KEY: misc_feature
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 90

ttcggctgtg aagagagtgt gcca 24

<210> SEQ ID NO 91
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<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
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<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 91

cgcttttgac acaagtggga ctgg 24

<210> SEQ ID NO 92
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 92

catagtgaca cccagaagct tcat 24

<210> SEQ ID NO 93
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<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
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<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 93

gagtcatccc tgcttcataa catt 24

<210> SEQ ID NO 94
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 94

gattaccaag tttcttcagc ttcc 24

<210> SEQ ID NO 95
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial

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<220> FEATURE:
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<220> FEATURE:
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 95

aggccttggc ccacttgacc acgt 24

<210> SEQ ID NO 96
<211> LENGTH: 24
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 96

agcatcctgg agttgacatt ggtg 24

<210> SEQ ID NO 97
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
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<220> FEATURE:
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<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 97

gacacactgg ctgtacatcc ggga 24

<210> SEQ ID NO 98
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<212> TYPE: DNA
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<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 98

gagccatcca aactcttgag agag 24

<210> SEQ ID NO 99
<211> LENGTH: 24
<212> TYPE: DNA
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<220> FEATURE:
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<220> FEATURE:
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<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 99

cagtgctttc atgcacagga attc 24

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<210> SEQ ID NO 100
<211> LENGTH: 24
<212> TYPE: DNA
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<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 100

attcgaagtt catcaaagaa tttt 24

<210> SEQ ID NO 101
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<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 101

atcgagttcc ttgatgtagt tcat 24

<210> SEQ ID NO 102
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
<223> OTHER INFORMATION: LNA oligomer Sequence/oligomer Sequence motif
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 102

gcacttgcac agagatgatc tctg 24

<210> SEQ ID NO 103
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<212> TYPE: DNA
<213> ORGANISM: artificial
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<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 103

aatagatggg cttgactttc ccag 24

<210> SEQ ID NO 104
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<212> TYPE: DNA
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<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

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<400> SEQUENCE: 104

ataacaggca gaagacatct gaaa 24

<210> SEQ ID NO 105
<211> LENGTH: 24
<212> TYPE: DNA
<213> ORGANISM: artificial
<220> FEATURE:
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<220> FEATURE:
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<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 105

attccccaag gcactgcaga ggag 24

<210> SEQ ID NO 106
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<212> TYPE: DNA
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<222> LOCATION: (1)..(24)
<223> OTHER INFORMATION: nucleotide or nucleotide analogues - optionally phosphorothioate

<400> SEQUENCE: 106

atgggctgac attcatagcc ttca 24

We claim:

1. An oligomer consisting of the formula:

$5'\text{-}{}^{Me}C_s{}^{Me}C_s{}^{Me}C_sa_sa_sg_sg_sc_sa_sc_st_sg_sc_sA_sG_sA\text{-}3'$ (SEQ ID NO: 77),

wherein uppercase letters denote beta-D-oxy-LNA monomers and lowercase letters denote DNA monomers, the subscript "s" denotes a phosphorothioate linkage, and ^{Me}C denotes a beta-D-oxy-LNA monomer containing a 5-methylcytosine base.

2. A conjugate comprising the oligomer of claim **1**, covalently attached to at least one moiety that is not a nucleic acid or a monomer.

3. A pharmaceutical composition comprising:

the oligomer of claim **1** or a conjugate comprising said oligomer covalently attached to at least one moiety that is not a nucleic acid or a monomer; and

a pharmaceutically acceptable diluent, carrier, salt or adjuvant.

4. A method of inhibiting the expression of androgen receptor in a cell, comprising:

contacting said cell with an effective amount of the oligomer of claim **1**.

5. A method of inhibiting the expression of androgen receptor in a cell, comprising:

contacting said cell with an effective amount of a conjugate according to claim **2**.

6. A method of inhibiting the expression of androgen receptor in a tissue of a mammal, comprising:

contacting said tissue with an effective amount of the oligomer of claim **1**.

7. A method of inhibiting the expression of androgen receptor in a tissue of a mammal comprising:

contacting said tissue with an effective amount of a conjugate according to claim **2**.

8. A method of inhibiting the expression of an androgen receptor target gene in a cell or tissue of a mammal, comprising:

contacting said cell or tissue with an effective amount of the oligomer of claim **1**.

9. A method of treating a cancer in a mammal comprising administering to said mammal an effective amount of the oligomer of claim **1**, wherein the cancer is selected from the group consisting of breast cancer and prostate cancer.

10. An activated oligomer comprising:

the oligomer of claim **1**; and

at least one functional group covalently attached thereto at one or more positions independently selected from the 5'-end, the 3' end, the 2'-OH of a ribose sugar, and the base.

* * * * *